The Diagnosis and Management

of

Primary Root Caries

by

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ABSTRACT

Dentine caries has a microbiological aetiology whilst the diagnosis relies on clinical signs. This study examined relationships between the Locations, Colours, Textures, Sizes, Perceived Treatment Needs, Cavitations and microbiological characteristics of Primary Root Caries. The relationships between some microflora of Primary Root Caries deemed to require restoration and the overlying plaque as well as the effects of a Chemotherapeutic agent on some microflora of lesions were also studied. In all, 610 lesions in 303 patients were investigated.

447 lesions in 169 patients were examined in the main study. The Locations of lesions were related to the gingival margins; Colours were designated: Black, Yellow, Light Brown or Dark Brown; Textures were recorded as Hard, Leathery or Soft, Sizes as products of Heights and Widths, and Cavitations as the greatest loss of surface contour. The total numbers of colony forming units ; Gram-positive pleomorphic rods; Mutans streptococci; Lactobacilli; and Yeasts expressed as Log_{10} as well as the proportions and Frequency of isolation, in each dentine biopsy were determined, eg

- 90.9 percent of Soft but only 3.3 percent of Hard lesions were <1 mm from gingivae ($P < 0.001$);
- 68.6 percent of Soft but only 6.7 percent of Hard lesions were sized >7 mm^2 ($P < 0.01$);
- more cavitated lesions were larger ($P < 0.01$);

- higher total numbers, proportions and frequencies of isolation of Mutans streptococci and Lactobacilli were in Soft and Leathery than in Hard lesions ($P < 0.01$);
- the mean total numbers of colony forming units, Mutans streptococci, Lactobacilli and Gram-positive pleomorphic rods were less in each group of lesions with a reduced Perceived Treatment Need ($P < 0.01$);
- lesions deemed to require restoration most frequently contained Yeasts ($P < 0.01$);
- the most reliable indicators of microbiological activity were Texture and Location rather than Colour;

From 81 Primary Root Caries lesions deemed to require restoration in 52 patients, not amongst the 169 in the larger study, higher proportions of Gram-positive pleomorphic rods ($P < 0.001$) and Lactobacilli ($P < 0.01$) were in carious dentine than in the associated plaque, indicating that precision in sampling is paramount. 42 of 82 lesions deemed to require restoration in another 82 patients, were coated with a varnish containing 1 weight (wt) percent chlorhexidine and 1 wt percent thymol and after 24 hours these and the lesions not varnished were biopsied. The varnish significantly reduced the numbers of micro-organisms (total colony forming units, $P < 0.001$; Mutans streptococci, Lactobacilli and Yeasts, $P < 0.01$).

These studies will help clinicians and epidemiologists to diagnose the levels of activity in Primary Root Caries and to indicate how Chemotherapy rather than the removal of carious dentine might be developed as a preferred strategy for its management.

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I INTRODUCTION

The teeth of human dentitions are composed of three tissues: the pulp-dentine complex which is protected by the enamel of the anatomical crowns; and on the surface of the anatomical roots it is covered by a thin layer of cementum, through which attachments to the alveolar bone and the gingivae are maintained by means of the periodontal ligament. Though these tissues may be lost through acute trauma, acid erosion, toothbrush abrasion, or attrition, the great destructive disease of teeth is dental caries, which may be defined as the acid dissolution of enamel, dentine or cementum as a consequence of the metabolism of micro-organisms living within deposits on the teeth known as plaque.

Traditionally, dental caries was considered to be mainly a disease of the young, attacking only the orally exposed parts of teeth: the anatomical crowns, so the primary attack was on enamel, for neither of the other two tissues was accessible. Enamel is a secretion rather than a true tissue, for no viable ameloblasts exist after the eruption of the teeth, and therefore, though enamel caries involves phases of acid dissolution followed by some recalcification, the body is incapable of positive protective action. Nevertheless, enamel caries is a relatively slow process compared with caries of the dentine; once this tissue is breached, its destruction leads to the need for restorative dental care, to the possible involvement of the pulp itself, and not infrequently to the loss of the tooth affected. When it became proven that the incorporation of fluoride ions into the hydroxy-apatite lattice of enamel significantly enhanced its resistance to caries and action was taken to ensure that this occurred, either through the ingestion of fluoride through the water supply and sometimes in tablet form, or through its

adsorption from toothpastes or mouthwashes, the early destruction of coronal enamel and then dentine abated and the foundation was laid for more teeth to be retained into middle and old age. This process was also enhanced by an increasing awareness amongst, not only the dental profession, but also the population as a whole, of the beneficial roles of fissure sealing, improved diets and oral hygiene in the prevention of caries and, as a result, a wider appreciation that the loss of teeth was not inevitable. Coupled with this has been a shift in the demography of society as a whole towards there being a greater proportion of older people than in earlier generations (Todd and Lader, 1991). So, today, there are more older people with more teeth potentially subject to dental disease than ever before.

The improvement in the dental health of UK adults was reported by Downer (1991) as a result of data collected from the UK dental surveys from 1968-1988. This showed that whilst 63 percent of the adult population in England and Wales were dentate in 1968, this proportion rose to 70 percent in 1978 and 79 percent in the 1988 survey. It was calculated that the proportion of the UK population with no natural teeth would fall to 14 percent in 1998, 10 percent in 2008 and 6 percent in 2028. It was predicted that by 2008, 65 percent of the over 65 year olds would retain some natural teeth, this proportion rising to 85 percent by 2038. There was an overall reduction in the mean DMFT (all ages) from 19.2 (1968) to 17.0 (1988). Whilst this decline was substantial for young adults aged 16-24 in England and Wales, ie 15.6 (1968) to 10.4 (1988), for older adults the reduction was, not surprisingly, more modest; 23.1 (1968) to 22.5 (1988) for 55 year olds and over.

The 1988 survey presented for the first time information about root caries, the highest prevalence being found in the 55-64 year old group with a mean of 0.7 teeth having root caries, and 1.2 having had restorations placed in their roots. Downer (1991) states that at the upper end of the age spectrum, an increasing proportion will be expected to remain substantially dentate.

The second major group of dental diseases affects the supporting structures of the teeth: the periodontal ligament and the alveolar bone. Periodontal diseases, as caries, are usually, though not always, very slow destructive processes which result in the loss of the attachments between teeth and bone through the cementum, and the loss of bone itself. As a consequence, not only the enamel of the crown but also the cementum of the root become exposed to the oral environment. In marked contrast to enamel, cementum is only moderately mineralised, thin, and porous. It is, therefore, incapable of providing significant protection to root-dentine from succumbing to dental caries and, because the cementum is so thin, it is in fact impossible to distinguish clinically between caries affecting one or both tissues, and not appropriate, therefore, to describe the condition as 'cemental caries' as did Hix and O'Leary (1976).

Though the crowns of teeth have been defined in two ways: the anatomical crown being that part of the tooth covered by enamel and the clinical crown being that part of the tooth within the oral cavity; the distinction is confusing and it is now customary to accept that the crown means the anatomical crown, therefore, the root is the anatomical root; the junction between the two being the cemento-enamel junction. Clearly, the dentine of the root may become carious either as a result of caries commencing on the

root surface itself or as a consequence of the extension of a carious lesion in coronal dentine which had been initiated as a sequel to caries in enamel. This has aggravated a confusion in terminology when reference is made to caries commencing on the surfaces of the roots of teeth.

Lowenthal (1967) called it 'Atypical', now clearly inappropriate if only in view of its prevalence; Schamschula et al (1972): 'Root Surface Caries' implying that the lesions had no depth; Hix and O'Leary (1976): 'Cemental Caries', but caries of the cementum is not clinically detectable (Mellberg, 1986); Hecht and Friedman (1949): 'Cervical Caries', but the cervical regions of teeth include the gingival third of the crown; whilst Jordan and Sumney (1973) use the terms 'Radicular', 'Erosion' and 'Senile', but erosion is by definition a different process to caries (Williams, 1987), senile implies that either the process or the patient is old, and radicular is a longer and more pedantic word than 'Root'.

The term 'Root Caries', used by Hazen et al (1973), has the advantage of being brief and accurate in describing carious lesions of the roots of teeth but it does not exclude root caries occurring as a sequence to coronal dentine caries. It is therefore desirable to refer to the former as 'Primary Root Caries' (Lynch, 1986) to avoid any confusion and it is with this condition that this work is concerned.

In societies which traditionally consume raw tough foods the teeth become severely attrited: cusp heights are reduced, even grooves are affected and the contact areas on the approximal surfaces are worn. Such diets are generally high in the complex carbohydrates found in grains, fruits and root vegetables but low in fermentable carbohydrates such as sucrose;

and much effort is involved in chewing with a high secretion of saliva. All these factors tend to reduce the chance of enamel caries becoming prevalent. Cultural changes which bring about the opposite circumstance in any society are therefore likely to increase the chance of enamel caries being initiated. Thus many authors (Leigh, 1925; Hardwick, 1960; Moore and Corbett, 1971, 1973) have reported that examinations of mediaeval and Anglo-Saxon skulls revealed relatively low incidences of coronal caries in the latter populations, but an all too familiar high prevalence in the former. However, they also noted that Anglo-Saxon populations experienced more Primary Root Caries than coronal caries, confirming the finding of Miles (1969). This presumably infers that the lack of plaque control resulted first in early periodontal disease and then in Primary Root Caries, possibly associated with the retention of complex carbohydrates for extensive periods on susceptible cementum and dentine as well as gingivae, which would support cariogenic micro-organisms adequately and for sufficient time to cause destruction.

The reduced impact of coronal caries on the dentitions of Western societies that has been referred to, mimics in many ways the situation found in Anglo-Saxon populations and has resulted in a growing interest over the past twenty years, among patients, clinicians, researchers, and the manufacturers of restorative materials and preventive agents, in Primary Root Caries.

The rise in interest in root caries was believed by Rozier and Beck (1991) to be due to the transition of dental caries from an almost universal disease of children and adolescents to a condition primarily occurring in high risk groups and individuals, the dramatic ageing of most industrialised

societies, the decrease in tooth loss and the implication of that decrease for older adults retaining more teeth, and the epidemiological finding that root caries was predominantly a disease of middle and old-age groups.

However, reliable criteria for the diagnosis of the disease and its management implications have neither been adequately developed or validated, indeed they have rarely been vigorously addressed (Beighton et al, 1993a). It would seem to be highly desirable in the interests of both sound patient management and of epidemiological data for reliable, fully validated methods of diagnosis to be established.

II BACKGROUND

The background to this study of Primary Root Caries that has been introduced will be provided with respect to its:

- Diagnosis and Epidemiology
- Aetiology
- Management

II-100 Diagnosis and Epidemiology

II-110 Introduction

Studies of Primary Root Caries involving microbiology, pathology, epidemiology or clinical trials have used very different definitions for classifying its diagnostic criteria, making comparisons between studies difficult or impossible, a fact noted by Rytomaa (1986). The criteria are neither sufficiently specific to permit the reliable identification of lesions; detailed enough to indicate the progression; nor do they provide adequate bases for treatment needs to be determined. Accurate sensitive diagnoses are required (Pitts, 1991c) and new diagnostic aids require ongoing evaluation (Pitts, 1992). There is clearly a need to standardise terminology and the definitions applied to the diagnosis of Primary Root Caries.

Hunt and Beck (1985) described the difficulties experienced in distinguishing between:

- one and two surface lesions;
- restored carious and restored abrasion lesions;

- Primary Root Caries proper and caries of root dentine consequent to caries of coronal dentine.

Recurrent Primary Root Caries has also posed a problem, Vehkalahti (1987c) in her study did not include such lesions whilst Hix and O'Leary (1976) did. In attempts to overcome such inconsistencies Hunt and Beck (1985) and Katz (1986) proposed the use of various conventions. Incipient lesions have been variously described as being of Soft, irregular Texture with no surface defects (Billing et al, 1985), or having Cavitation or softened areas (Hix and O'Leary, 1976); whilst active lesions have been defined as being at or near the cemento-enamel junction (Mount, 1986), and at or below the level of the free margin of the gingivae (Banting and Ellen, 1976). The progression of Primary Root Caries has been variously described as: tending to spread laterally, or without Cavitation (Fejerskov and Nyvad, 1986); and as ranging from having no surface defects to involving the pulp (Billings et al, 1985). Jensen and Kohout (1988) and De Paola et al (l989) attempted to modify the criteria that had been used up to that time to systematise the diagnostic criteria applicable to Primary Root Caries.

The diagnostic problems associated with Primary Root Caries have no more obvious consequences than in the field of epidemiology. These difficulties are compounded by a failure to agree on whether all the lesions diagnosed as Primary Root Caries should be included or only those deemed to be active. The most common practice has been to record only the latter which clearly must have resulted in an under-estimation of the true count and, since only root surfaces coronal to the gingival margin (excluding surfaces within periodontal pockets) are normally examined, the true figures will undoubtedly be even higher.

This lack of standard diagnostic criteria prevents realistic comparisons of different studies to be made, an issue presented by Aherne et al (1990). In three national surveys in the United Kingdom (Todd and Lader, 1991), the USA (Miller et al, 1987) and Ireland (O'Mullane et al, 1989), none of the protocols employed were comparable. Whilst Colour descriptions were the same, lesion shapes; the methods of probing; the procedures defined for lesions involving both the crowns and roots of teeth; and for taking account of the presence or absence of gingival recession, all varied. In the USA study (Miller et al, 1987), resistance to the withdrawal of a probe was taken to indicate sound dentine and cementum whilst the diagnosis of Primary Root Caries required the penetration of a probe and its easy removal from the tissue. In both the other surveys, as with most studies, a resistance to the withdrawal of a sharp probe was defined as indicating the presence of caries. Nor did the US survey distinguish between a tooth surface in which no root dentine was exposed and one in which sound root dentine was exposed as a consequence of gingival recession, whilst in both the UK and Irish surveys gingival recession was recorded as a distinct entity.

Other sources of potential inaccuracies in the epidemiological data that have been generated include a failure to clearly define when a lesion had developed in the root dentine, or whether it was an extension of a lesion which initiated in the cervical enamel of the crown (Beck, 1990); and relate the Location of a lesion to the gingival margin despite the findings that 80 percent of Soft lesions are found within 1 mm of it (Hellyer et al, 1990). Rarely has it been specified how far a lesion needs to have extended laterally before additional surfaces are judged to be involved, nor the criteria to be applied when attempting to distinguish between Primary Root Caries,

and abrasion lesions which have been restored. Even the presence or absence of Cavitation is not considered to be a matter of significance in most studies.

The considerable variations in the diagnostic criteria that have been referred to will not be repeated but one aspect which seems to have created particular problems in epidemiological studies has been the classification of Primary Root Caries and of restorations which are deemed to have been placed as a consequence of Primary Root Caries. Thus, De Paola et al (1989) reported on variations in the prevalence figures for Primary Root Caries in a study of 223 subjects aged between 44 and 64 years. Only 23 percent were judged to have been affected by one or more Primary Root Caries lesions, a proportion that increased to 85 percent when restorations involving parts of the crowns and roots were included along with any recurrent caries. Such dramatic changes in epidemiological data illustrate the need to improve the diagnostic basis for Primary Root Caries which would undoubtedly require the use and acceptance of additional diagnostic aids to those customarily employed. Some of these 'diagnostic' tools have been discussed and it was concluded that there was no one single method suitable for every situation (Pitts, 1991d).

II-120 Diagnosis

Visual, tactile, radiographic and locational have been considered to be appropriate criteria to be considered in the diagnosis of Primary Root Caries (Katz, 1984). Particular emphasis has usually been laid on the Colour and the Texture and the presence or absence of Cavitation, whilst the Locations

of lesions have not been incorporated into any diagnostic classification nor have symptoms nor any possible photographic recordings and analyses.

II-121 Symptoms - The first phase of any diagnostic exercise undertaken by wise clinicians is to listen to, then record and analyse the words of their patient, a procedure known as 'History Taking'. Frequently such histories include the patients' descriptions of any symptoms they are experiencing, though dentists invariably need to develop the art of extracting such information in a meaningful form. It is perhaps surprising, therefore, that symptoms arising from Primary Root Caries have never been incorporated into any systematic approach to its diagnosis. This may be due to the fact that symptoms are only rarely associated with root caries (Banting and Ellen, 1976; Katz, 1990). Since Primary Root Caries is essentially carious activity in dentine, symptoms are likely to be those characteristic of any affectation of the pulp-dentine complex. The stimulation of this tissue is the cause of what most patients would describe as 'tooth-ache' and routinely such symptoms are classified in accordance with well established characteristics including:

- the period of time since they first manifested;
- the nature of the stimulus that initiated them;
- their frequency;
- their duration;
- their spontaneity;
- the time they last presented;
- the nature of any actions that alleviate them;
- their Location;
- their onset in relation to some earlier action.

Whilst such symptoms can invariably indicate the presence of a condition which is stimulating an abnormal response from the pulp-dentine complex it does not, of course, differentiate between the many alternate causes of such stimulation, including Primary Root Caries. However, in relation to other criteria, notably the signs observed by clinicians, it might be both helpful and appropriate to consider the incorporation of any symptoms associated with Primary Root Caries into any overall diagnosis.

II-122 Clinical Signs - Visual indications of the presence of pathology such as Primary Root Caries are normally referred to as clinical signs. This is the most important group of factors on which diagnoses are made, especially when epidemiological evidence is being recorded and simple observation is likely to be the only means available. Three groups of criteria need to be considered:

- Location and Size. Banting and Courtright (1975) studied extracted teeth and concluded that Primary Root Caries always begins at or near to the cemento-enamel junction, but Fejerskov and Nyvad (1986) described them as commencing adjacent to the gingival margin, irrespective of whether or not other lesions existed on the same root surface though more coronally. The criteria defined by Katz (1984) are imprecise for they can be interpreted to mean either the position of a lesion on the root surface only or on the tooth as a whole.

Banting et al (1985) used both the gingival margin and the cemento-enamel junction as reference points and found 54 percent of new lesions to be at the cemento-enamel junction and within 8 mm of the gingival margin.

However, over this three year study only 8 percent of new lesions developed on root surfaces more than 2 mm from the gingival crest. In 1986, Ravald et al reported that, over an eight year period, 25 percent of new lesions were located at the cemento-enamel junction with only 7 percent at the gingival margin, but the majority of lesions diagnosed were recurrent.

It would seem that Primary Root Caries affects the approximal surfaces of teeth more commonly than either facial or lingual surfaces (Leigh, 1925; Hardwick, 1960; Corbett and Moore, 1971). Miles (1969) found this to be the case in his study of Anglo-Saxon skulls, as did Banting and Courtright (1975) and Westbrook et al (1974) in contemporary extracted teeth. However, epidemiological studies by Keltjens et al (1988) and Wallace et al (1988a) suggest that facial surfaces are more commonly affected whilst Sumney et al (1973) suggest that facial and lingual surfaces are most affected. It is possible that these discrepancies are due mainly to the differing natures of these groups of studies. Approximal surfaces are notoriously difficult to both see and to touch *in vivo*, whilst facial surfaces are by far the most accessible. It is, therefore, likely that in any epidemiological survey a far higher proportion of the latter will be diagnosed than lingually-located lesions and certainly more than approximal lesions.

A number of studies have included the categorisation of Primary Root Caries according to Size. Banting et al (1980) described lesions as discrete and well defined whilst others (Sumney et al, 1973; Lohse et al, 1977) depict them as ill-defined. Linear narrow lesions which encircle teeth close to the cemento-enamel junction were found in 9 percent of the extracted teeth examined by Westbrook et al (1974) whilst Banting and Courtright (1975) described similar lesions as linear and band-like in contrast to others which

were round, or elliptical. They also speculated that Primary Root Caries commences as a number of small, round, discrete lesions near the cemento-enamel junction which later coalesce into larger elongated lesions. Kidd (1990) held a similar view and described some lesions as being 'abandoned' near to the cemento-enamel junction by the receding gingivae, whilst quite new lesions can develop later at the level of the repositioned gingival crest.

Hellyer et al (1990) related the Size of Primary Root Caries lesions, ie the products of their maximum height and width, as measured with a graduated periodontal probe, with their Texture and found that the significantly larger lesions were softer than smaller ones. Clearly, the Size of a lesion has implications for treatment needs, in spite of which it has rarely been considered a factor in the diagnosis of Primary Root Caries. Thus, a shallow Soft or Leathery lesion less than 1 mm in diameter would undoubtedly be recorded as an actual lesion in any prevalence study but would not be categorised differently from a Soft lesion involving the whole of an exposed root surface from the cemento-enamel junction to the gingival margin though, clearly, the treatment implications would be dramatically different.

- Colour. In attempts to define Primary Root Caries several authors have used the term 'Discoloured', presumably this means in comparison to any neighbouring tissue deemed to be sound (Banting et al, 1980; Jensen and Kohout, 1988; and Katz, 1984), whilst others do not consider a change in Colour as essential to its diagnosis (Beck et al, 1985; Gustavsen et al, 1988; Hix and O'Leary, 1976; Lohse et al, 1977; Ravald and Hamp, 1981; and Vehkalahti, 1987b). Since Primary Root Caries, is by definition, caries of the root dentine (the cementum being very thin), then

Colour change must infer a change in the Colour of root dentine. The structure of dentine is complex and both its relatively high organic content and its tubular structure make it liable to take on varying hues. The simple fact that tubules and their peri-tubular matrices are more widely spaced in the more superficial layers of dentine than in the deeper ones; that tubule diameters are greater near to the pulp than those nearer the outer tooth surface; and that peri-tubular matrix can be more highly calcified than inter-tubular matrix, means that even sound mature dentine will vary in tone with depth. However, dentine Colour can be affected by many other factors.

First are the 'Extrinsic Stains' which can be removed with mildly abrasive pastes. They may originate from foods or drinks, notably tea, coffee and red wine; from certain drugs such as chlorhexidine used in mouth washes; or from tobacco or metabolic products of chromogenic bacteria. Clearly, as Katz (1990) advocated, these need to be removed if a colour change in root dentine is to be used with any confidence to diagnose Primary Root Caries.

Arguably one of the most significant conditions to take into account is toothbrush abrasion, for as a consequence, the deeper layers of root dentine are exposed. These have different tones and, not infrequently, dentinal tubules may be opened up enabling extrinsic staining to pass into the dentine itself and make it impossible to remove with an abrasive paste. Should trauma, or coronal caries result in pulp necrosis, the break-down products of haemoglobin in particular, which would produce a temporary bruise in soft tissue, seep into dentine from its pulpal surface and produce a permanent discoloration. Similarly, should a tooth be subject to 'Internal Resorption' the

vascular pulp will show through the translucent and thinner root dentine and give the latter a discoloured appearance.

Root dentine can appear discoloured from a number of intrinsic defects in the structure of dentine (Kidd, 1983): for example, dentinogenesis imperfecta; hereditary porphyria; prenatal or neonatal disturbances; haemolytic disease of the new born; febrile illness; rickets; epidermolysis bullosa dystrophia; or fluorosis. Some years ago, young children provided with the tetracycline drugs during the development of the permanent dentition could suffer from severe permanent discoloration of the dentine as a consequence.

Finally, a number of substances used by dentists can give rise to the staining of root dentine. Perhaps the commonest is silver amalgam, especially the older alloys which tended to corrode far more than the modern ones. Silver and copper salts which are products of corrosion seep into the adjacent dentine and discolour it. Ironically, these same salts tend to inhibit the growth of the organisms which give rise to caries, so dentine discoloured in this way is less likely to be carious than otherwise.

The semantics of the Colours ascribed to altered root dentine are many and imprecise as illustrated in Table II-122a. However, it needs to be appreciated that most authors listed ascribe the type of lesion not only on a basis of Colour but also on its Texture and/or any Cavitation exhibited.

Colour	Caries State Ascribed	Authors
Yellowish	Active	Nyvad & Fejerskov 1982, 1986
	Active	Nordenram et al 1988
Yellow/orange	Active	Miller et al 1987
	Active	Todd and Lader 1991
Yellowish/light brown	Active	Banting and Ellen 1976
	Active	Hix & O'Leary 1976
	Active	Ravald and Hamp 1981
	Incipient or active	De Paola et al 1989a, b
	Active	Ravald & Birkhed 1991
	Active	Fejerskov et al 1991
	Active	Adriaens and Nyvad 1992
	Active	Eliasson et al 1992
Light brown	Active	Nyvad & Fejerskov 1982, 1986
	Active	Fejerskov et al 1991
Light tan to brown	Incipient	Billings et al 1985
Tan	Active	Todd and Lader 1991
Tan to dark brown	Active/shallow	Billings et al 1985
Light brown to dark brown	Active/cavitated	Billings et al 1985
	Active	Leske and Ripa 1989a, b
	Inactive	Adriaens & Nyvad 1992
Brownish	Active	Nordenram et al 1988
Brown/dark brown	Active/pulpal involvement	Billings et al 1985
Dark brown	Inactive	Fejerskov et al 1991
	Inactive	Eliasson et al 1992
Dark Brown Almost Black	Passive/Remissive	Nyvad & Fejerskov 1982
Black	Passive/remissive	Banting & Ellen 1976
	Chronic carious lesion	Nordenram et al 1988
	Arrested	De Paola et al 1989a, b
	Inactive	Fejerskov et al 1991
	Passive	Todd & Lader 1991
	Inactive	Adriaens & Nyvad 1992
	Inactive	Eliasson et al 1992
Discoloured	Active*	Banting et al 1980
	Active/Inactive	Katz 1984
	Active	Jensen and Kohout 1988
	Active	Bauer et al 1988
Pigmented	Arrested	Massler 1980
No specific colour noted	No Activity Specified	Lohse et al 1977
		Beck et al 1985
		Vehkalahti 1987c
		Gustavsen et al 1988

*Suggested rather than stated activity

Table II-122a: The Colours used to describe root caries in the literature and the disease states ascribed to them

With the exception of Nordenram et al (1988) every author listed considered Black/Dark Brown lesions to be in a passive, arrested, or remissive state. Nyvad and Fejerskov (1982) described Primary Root Caries as one or more well defined, discoloured areas located, predominantly, along the cemento-enamel junction. Yellowish or Light Brown softened dentine not exhibiting any surface Cavitation they termed 'Active'; whilst Dark Brown or Black root dentine they termed 'Passive', though their Texture could be softer than sound dentine but more Leathery than active lesions (frequently as Hard as the sound tissue).

Simplified criteria for diagnosing Primary Root Caries which it was claimed could be used in either epidemiological studies or in clinical practice have been described by Nyvad and Fejerskov (1986). 'Active Lesions' they describe as well-defined and Yellow or Light Brown in Colour; they may be covered by visible plaque and/or on probing have a softened or Leathery consistency. 'Inactive Lesions', on the other hand, were described as well defined and Dark Brown or Black with smooth, shiny surfaces which were Hard to probing using moderate pressure. Though these authors were undoubtedly correct that it is essential to distinguish between active and inactive caries, they so rigidly related the Colour of any lesion to its level of activity that even root surfaces which were Soft to the probe were not classified as being carious as long as they were Black. The need for epidemiologists to diagnose and classify Primary Root Caries preferably by vision alone, gives rise to considerable problems and inconsistencies. Even the need to remove extrinsic stains by polishing tooth surfaces with mildly abrasive polishing pastes before reliable diagnosis on visual appearance alone can be made, poses difficulties, as stated by Katz (1990). These are

evident from the basis of diagnosis used in the UK Oral Health Survey (Todd and Lader, 1991) in which Yellow/orange, tan, or Light Brown root surfaces were diagnosed as exhibiting active carious lesions, whilst dark or almost Black surfaces were designated as arrested lesions.

Hellyer et al (1990) considered that the same Colour range from Yellow to Black could be exhibited by both Hard and Soft Primary Root Caries and, therefore, found no correlation between their Colour and their Texture. This makes it difficult to conclude that Colour alone can be a reliable indicator of the activity of any lesion.

Using a rather simplistic approach to the problem by studying only extracted teeth exhibiting Primary Root Caries, Nordenram et al (1988) proposed the following classification in which, once again, rigid Colour:Texture combinations were used as the basis of classification:

- 'Active' being either Yellow or Brown, and Soft were sub-divided into those without and those with loss of dentine;

- 'Chronic' being Black/Brown, Hard and shiny.

De Paola et al (l989a, b) significantly extended the number of designated groups of root surface lesions to nine as illustrated in Table II-122b. However, no mention is made of any pre-examination removal of extrinsic stains in this study and the classification has all the rigidity of the classifications previously referred to.

Code	Designation	Description
C1	Incipient lesion	A well defined softened area, yellowish or light brown in colour, but without cavitation upon initial inspection, ie the morphological integrity of the surface is undisturbed before probing.
C2	Frank cavitation	A softened area, yellowish or light brown in colour, with a disruption of the normal surface contour, ie there is a discontinuity or break in the surface, even prior to probing.
AC	Arrested caries	A darkly stained, almost black area with a leathery or hardened consistency so that penetration by the explorer is difficult and there is no resistance to withdrawal; there may or may not be cavitation of the surface.
CF	Secondary caries	Caries at the margin of a restoration as evidenced by a yellowish brown softening at the interface of the restoration and the root surface.
F	Root restoration	A restorative material which has been inserted on the root surface only.
R1	Overlapping lesion	A lesion or restoration which is primarily on the enamel but extends on to the root.
R2	Overlapping lesion	A lesion or restoration which is primarily on or over the root but extends on to the enamel.
A	Root abrasion	A wedge-shaped defect, softly angled in the early stage, sharply-angled in later stages, with highly polished exposed dentine.
E	Root erosion*	A shallow, broad, smooth disc-like depression resulting from a chemical process. (*Root erosion has been observed in only one subject to date.)

Table II-122b: Clinical definitions used for the assessment of root caries after De Paola et al (1989)

The problems incurred in using only Colour change to diagnose Primary Root Caries were highlighted by Bader et al (1993) who provided a group of general dental practitioners with three colour photographs of non-carious root surfaces and a fourth of an active carious lesion. The majority of these dentists are reported as indicating that, on visual appearance alone they would have restored the non-carious root surfaces. However, the authors do not indicate the basis on which the five teeth were diagnosed as being either carious or non-carious.

It is difficult not to agree, from the evidence published to date, that Katz (1986) was correct in concluding that any assumption that the discoloration of root surfaces is caused only by caries and no other cause must be unsound and, until such time as it becomes possible to exclude other aetiology, Colour change should be omitted from the criteria employed to judge the level of carious activity in root dentine.

- Surface Contour. The loss of dentine from the surfaces of teeth can be observed without instrumentation being resorted to, though the use of a probe to enhance visual judgement by tactile sensation is common. It is not surprising, therefore, to find that the loss of dentine has been employed and reported by a number of researchers. Changes have been described as 'Cavitation'; 'Surface Defects'; or 'Loss of Surface Continuity'. Sumney et al (1973) in fact incorporated Cavitation into their definition of Primary Root Caries viz 'Cavitations below the cemento-enamel junction', whilst many have stated that such lesions may be cavitated (Nyvad and Fejerskov, 1982; Fejerskov and Nyvad, 1986; Gustavsen et al, 1988; and Jensen and Kohout, 1988). Leske and Ripa (1989) used the term "loss of surface continuity" as a visual criterion for the diagnosis of active root caries.

Katz (1984) was of the opinion that all cavitated lesions are active, but by 1986 he had modified this, suggesting that cavitated lesions which are not Soft are inactive. Earlier than this Hix and O'Leary (1976) defined active Primary Root caries as 'a cavitation or softened area of the root surface' and reported that a study of extracted teeth revealed that Cavitation varied from a slight surface etching to a cavity of 3 mm in depth; but that only 3 percent of lesions had progressed to a depth sufficient to involve the pulp.

Katz (1984) divided Primary Root Caries into two main categories each with two sub-divisions, but this was done purely on appearance without any apparent logical attempt to indicate what these categories inferred:

- lesions exhibiting gross Cavitation and either a darkened or discoloured appearance or a tacky or Leathery feel upon probing with a moderate pressure;

- lesions without gross Cavitation but with a darkened, discoloured appearance which may or may not have a tacky or Leathery feel upon probing with a moderate pressure.

The first category seems to suggest that a cavitated lesion may be discoloured and Hard or not discoloured but Soft, whilst the second indicates that a non-cavitated lesion will be discoloured but may be either Hard or Soft. The inference is only that the non-cavitated Hard lesions are inactive, the others, presumably being active but the possibility of a different aetiology for discoloured intact surfaces is not clarified.

In his later paper, Katz (1986) equated discoloured, Hard, non-cavitated lesions with arrested caries, and therefore suggested that this should be ignored. He noted that non-cavitated Primary Root Caries treated by Chemotherapeutic agents may later present only discoloured intact and Hard surfaces and should then not be scored as carious. He then went on to propose visual:tactile criteria for identifying Primary Root Caries which may occur on any root surface:

> 'Active Lesions' have a darkened, discoloured appearance and a tacky, Leathery feel upon probing with moderate pressure. They may or may not be cavitated;
>
> 'Inactive Lesions' may have a darkened, discoloured appearance and may be cavitated but they do not have a tacky, Leathery feel on probing.

It would seem, therefore, that in Katz's opinion the basic characteristic differentiating active from inactive lesions is Texture, he does not attempt to distinguish between the various aetiological processes which cause the discoloration of dentine, any more than he faces the more complex differentiation suggested a year earlier by Billings et al (1985). The latter researchers described four grades of severity for Primary Root Caries:

> 'Grade 1 - Incipient Lesions' have a Soft, irregular Texture which can be penetrated with an explorer but no surface defect, the pigmentation varying from Light Tan to Brown;

'Grade 2 - Shallow Lesions' have a Soft, irregular, rough Texture which can be penetrated with a dental explorer and surface defects less than 0.5 mm deep, the pigmentation varying from tan to Dark Brown;

'Grade 3 - Cavitation Lesions' have a Soft Texture which can be penetrated with a dental explorer and surface defects more than 0.5 mm deep but without pulpal involvement, the pigmentation varies from Light Brown to Dark Brown;

'Grade 4 - Pulpal Lesions' are deeply penetrating with pulpal involvement, the pigmentation varies from Brown to Dark Brown.

It would seem, therefore, that this somewhat lengthy description incorporates only a small number of simple facts:

- caries diagnosis is made on Texture, ie it is softer than sound dentine, a fact that all clinicians are aware of;
- the Colour of a carious lesion may vary from Light Brown to Dark Brown but its Colour does not relate directly to its activity, more to its age;
- a lesion may exhibit either no loss of tissue or an extensive amount.

Clearly, this paper fails to carry forward any real solution to the problem of categorising Primary Root Caries. This is, perhaps not surprising

since only six patients were used as the sample on which the proposals were based.

The loss of dentine from the surfaces of roots is an important entity in their diagnosis and especially in the implications for treatment. It undoubtedly suggests that there may have been significant caries activity at some time, though erosion and abrasion may have been the main agents. It cannot be considered as a reliable indication of current caries activity, whether or not the dentine is also discoloured.

II - 123 Clinical Investigations. Under this heading need to be considered the actions which clinicians might take in the light of the symptoms reported by patients and any observations made. There are three groups: the Stimulation of the pulp-dentine; the Texture of a suspected lesion; and Radiographic characteristics.

- Stimulation of the Pulp-Dentine. As referred to above, patients commonly describe certain symptoms to dentists which are related to the presence of Primary Root Caries. However, because of the nature of the innervation of the pulp-dentine, as distinct from that of the periodontal ligament, patients are unable precisely to locate the source of the pain. The stimulation of the pulp-dentine of one tooth is commonly referred to other teeth in the same quadrant of the dentition and, not infrequently, to the other quadrant on the same side. Only very rarely indeed is it referred to the opposite side. If, therefore, symptoms may be arising from Primary Root Caries, it becomes an early priority to attempt to identify the particular tooth which is the source of the pain.

The nature and differential diagnosis of pulpal pain is a complex problem which tends to be based as much in art as science and it is inappropriate here to consider it in any detail. Suffice it to say that, whilst tooth movement, through biting or percussion, stimulates receptors in the bone and periodontium, it does not stimulate the pulp-dentine. This is achieved by bringing about a temperature change, by a change in the osmotic pressure of oral fluids or by touch. By far the most convenient and effective clinical methods are to cool individual teeth in sequence by means of a small pledget of cotton wool soaked and frosted with ethyl chloride or to touch a questionable surface. Whilst all vital teeth are likely to respond to a greater or lesser extent to being cooled down in this way, teeth with exposed root dentine and those with carious dentine of either the crown or root are more likely to produce very much more intense pain than would otherwise be the case. There can be other causes such as unlined metallic restorations and cracked or fractured teeth; but exposed roots, the essential precursor of Primary Root Caries and erosion or abrasion which need to be differentially diagnosed from it, are important elements.

The stimulation of the pulp-dentine as a factor in diagnosing Primary Root Caries is probably of value in only two areas: to help in identifying root surfaces at risk of developing lesions; and to provide some indication of the degree to which the pulp itself is responding to the stimulation of a lesion, which may infer its depth and the possibility of irreversible pulpal involvement. The greatest single problem attached to the use of pulp-dentine stimulation is the fact that the simple exposure of the root surface to the oral environment, without there being any caries, abrasion or erosion of that tissue can very commonly result in that surface being extremely

sensitive to any stimulation whether it be by touch, temperature change, or osmotic change brought about by sugar or salt.

- Texture of Lesions. Reference has already been made to the significance of the Texture of carious dentine in the diagnosis of Primary Root Caries, for it is difficult to separate it entirely from other characteristics. However, its importance certainly warrants its consideration as an individual entity (see Table II-123). Descriptions such as 'a tacky Leathery feel upon probing with moderate pressure' were used by Katz (1984) and by Nyvad and Fejerskov (1986), and ten years earlier, Hix and O'Leary (1976) advocated the use of a Hartzel No 3 explorer which could easily be inserted into carious dentine with moderate finger pressure. Newitter et al (1985) went further by proposing the use of a modified explorer (probe) to detect Primary Root Caries. They postulated that carious enamel is distinguished from sound enamel by the tactile difference perceived by the clinician. If, an explorer (probe) were to be modified by making its arm (shaft) flexible and its tip was bent at an angle of 30° to its long axis, then it would penetrate Primary Root Caries, bending the arm in the process, as a consequence the tip would be withdrawn at an angle to its long axis and tactile sensation would be enhanced. The device was tested by six blind-folded clinicians and an increased diagnostic sensitivity was confirmed, but as Kidd and Joyston-Bechal (1987) noted, the probing of suspicious lesions to identify 'stickiness' depends as much on the sharpness of the probe and the force exerted as anything else. Furthermore, an incipient lesion which may be managed conservatively through changes in plaque control and diet might well be adversely affected by any clinician creating a narrow defect in its surface by probing.

Texture	Disease Status	Description/Method	Author
Soft	Active	Explorer (Hartzel No 3) easily inserted with moderate finger pressure	Hix & O'Leary 1976 Ravald & Hamp 1981
	Active	Resistance to withdrawal of probe	Banting et al 1980
	Active	Softened dentine	Nyvad & Fejerskov 1982
	Active Active* Active*	Lesion penetrated with a dental explorer Lesion penetrated with a dental explorer Lesion penetrated with a dental explorer	Billings et al 1985 Beck et al 1985 Leske & Ripa 1989
	Active	Soft, spongy or greasy	Katz 1986 Nyvad & Fejerskov 1986
	Active	Explorer penetrates the tissue but can be removed easily	Miller et al 1987
	Active*	Softer than surrounding tissue	Gustavsen et al 1988
	Active	Resistance to withdrawal of a No 23 probe	Jensen & Kohout 1988
	Active	Softened area but not cavitated	De Paola et al 1989 a, b
	Active	Spongy	Hellyer et al 1990
	Active	Soft on probing with moderate pressure	Fejerskov et al 1991
Leathery	Passive or Remissive	Softer than surrounding cementum but more leathery than the active lesions	Nyvad & Fejerskov 1982
	Active	Tacky or leathery upon probing with moderate pressure with or without gross cavitations	Katz 1984
	Active	Tacky or leathery upon probing with moderate pressure with or without cavitation	Katz 1986
	Arrested	Leathery or hardened consistency so that penetration with an explorer is difficult and with no resistance to withdrawal	De Paola et al 1989a, b Banting & Ellen 1976 Fejerskov & Nyvad 1986 Katz 1984, 1986
	Active	Resistance to withdrawal of a No 3 Ash probe	Hellyer et al 1990
Hard	Inactive/ Arrested	Hard lesions may be harder than surrounding non-diseased root surface	Nyvad & Fejerskov 1982
	Arrested	Explorer did not easily penetrate the lesion and no resistance to withdrawal or hard/smooth/polished	Billings et al 1985
	Inactive	Smooth, shiny, hard to probe using moderate pressure	Fejerskov & Nyvad 1986 Katz 1984, 1986 Fejerskov et al 1991
	'Chronic' lesion	Hard and shiny	Nordenram et al 1988
	Arrested	Hard but sometimes leathery, penetration was difficult with an explorer, but no resistance to withdrawal	De Paola et al 1989a, b

*suggested rather than stated activity

Table II-123: The Textures of Lesions used to describe root caries in the literature and the description or method ascribed to each

To probe a doubtful lesion may not prove to be the best advice (Kidd, 1984; Pitts, 1991a), however, generally the 'careful' use of a probe may be helpful (Pitts, 1991b). Schaeken et al (1991a) used spoon excavators to diagnose the softness of Primary Root Caries, a procedure which must have been far more invasive than using a probe and, therefore, potentially even more damaging.

There would seem to be two basic methods for diagnosing Primary Root Caries on the basis of its Texture: either by description or by comparison with surrounding dentine deemed to be non-carious. As well as the 'tacky Leathery feel' described by Katz (1984) and others, terms such as 'soft', 'spongy', or 'greasy' have also been used. Indeed, both Katz (1986) and Nyvad and Fejerskov (1986) used them to define two categories of lesions, ie soft, spongy or greasy on the one hand or Leathery on the other. Hellyer et al (1990) classified lesions using both descriptions and comparisons eg Hard (as the surrounding dentine); Soft (spongy); or Leathery (resisted the withdrawal of an Ash No 3 probe); and they presumed both Leathery and Soft dentine to be actively carious.

These criteria of Texture would seem to be the most widely accepted ones for diagnosing active Primary Root Caries but, clearly, it is remarkably difficult to define or describe with any precision. The available methods for assessing Texture tend to be both imprecise and potentially damaging. Nyvad and Fejerskov (1987c), having shown that minerals can be taken up from saliva into the surface layers of Primary Root Caries, hypothesised that the probing of such lesions, in that it damages this layer, must be contra-indicated, though their own clinical evidence (Nyvad and Fejerskov, 1986)

seems to be weakened by their definitions of active and inactive Primary Root Caries.

In spite of all these problems, it would seem that Texture is the one clinical criterion which enables active Primary Root Caries to be distinguished from lesions caused by erosion, abrasion or other trauma and from lesions caused by a carious process which is no longer active. Furthermore, there are circumstances in which tactile investigations are the only means of identifying lesions, including those on the approximal surfaces of roots or within deep periodontal pockets, when diagnosis on the basis of vision alone would be impossible (Burt et al, 1986 and Locker et al, 1989).

When deeper lesions are involved, it is important to appreciate that circumpulpal dentine and peripheral dentine, even of the roots of teeth do have different morphologies. The concentration of large dentinal tubules, usually lacking any hypercalcified peritubular matrix, results in a tissue which is significantly softer, even when quite unaffected by the carious process, than peripheral dentine with its more widely spaced, small tubules, usually surrounded by highly mineralised peri-tubular matrix. Consequently, any judgement on Texture must make allowance for the depth of a lesion and of the type of dentine being assessed.

- Radiography. An invaluable aid to the diagnosis of approximal coronal enamel and dentine approximal caries is radiography, but to what extent is it helpful in the diagnosis of Primary Root Caries? Banting (1986) considered that it might assist with approximal lesions and that with the careful assessment of radiographs, the correct diagnosis can be made. He believed that approximal Primary Root Caries can be distinguished from

'cervical burnout' produced as a consequence of the surface contour of the teeth, by identifying what he described as an enamel 'Spine' or 'Stalactite' adjacent to the area of radiolucent dentine in the former. However, extreme care is necessary in obtaining, processing, magnifying and reading such radiographs (Pitts, 1991b).

Sumney et al (1973) identified an additional 14 approximal lesions from studying radiographs of a population of 30-60 year old individuals. This represented 8.5 percent of the total. However, Vehkalahti (1987a) considered that the use of radiography in any survey may well exaggerate the number of lesions of Primary Root Caries in view of the difficulty of excluding normal cervical radiolucency. This opinion was supported by the findings of Nordenram et al (1988) who found a 24 percent disagreement between visual and radiographic examinations in the diagnosis of Primary Root Caries on approximal surfaces. In this study using extracted teeth, surfaces diagnosed as sound on visual examination were diagnosed as carious when radiographs of them were studied. On the other hand, five obviously carious surfaces were diagnosed as sound from their radiographs.

It would certainly appear that radiographic evidence alone is an uncertain basis on which either the presence or absence of Primary Root Caries on the approximal surfaces of teeth can be diagnosed and the literature provides no evidence that radiography is of any value in confirming the characteristics of facial or lingual lesions.

II -124 Discussion and Conclusions. It would seem that a universally acceptable set of diagnostic criteria has proved to be unattainable up to the present time. Six years ago Nordenram et al (1988) drew the same

conclusion and no significant advance has been made since. Perhaps this is not surprising in view of the frequency with which rather confused logic seems to have been applied. For example, Mount (1986) stated that: 'A differential diagnosis between root surface caries and caries on the root surface is necessary.' Clearly such a statement implies that two different processes are underway or two different pathological conditions exist, and yet neither this author, nor others have reported that two different microbiological aetiological characteristics or two different sets of morphological changes in the dentine lesions are recognisable.

In 1988, Gustavsen et al published the results of a study of Primary Root Caries which was in many ways comparable to other studies and yet the diagnostic data were based on the dentists involved using only their clinical judgement when scoring, rather than attempting to apply detailed criteria defined by the others. It is, therefore, pertinent to ask if it is a matter of any significance that definitions vary, for it is possible or even probable that comparable conditions are being recorded in dissimilar ways by different workers (Hellyer and Lynch, 1990). In spite of this, it is unlikely that significant further progress can be made in the understanding of the disease process involved in Primary Root Caries unless a clear consensus is reached on the diagnostic criteria to be used in the definitions that are appropriate.

II-130 Prevalence and Incidence

II-131 Prevalence. The proportions of populations affected by Primary Root Caries (its prevalence) have been estimated, by a number of researchers studying different groups (Hellyer and Lynch, 1989a). Table II-131a records some overall figures from 23 studies. It would appear that 15-44 percent of the healthy ambulant, urban populations studied have some Primary Root Caries but the proportions dramatically increased amongst the institutionalised; the chronically sick; and the elderly, here in the latter rising to 89 percent. However, such figures need to be treated with caution and can be considered to be only crude estimates for, not only is it impossible to make direct comparisons between different studies in view of the many reasons already discussed, but the groups and teeth studied include both roots that were vulnerable due to gingival recession and roots that were not, because they were not exposed to the oral environment. Katz (1980) defined the Root Caries Index (RCI) in an attempt to define with greater accuracy the 'true attack rate', since it identifies the proportions of susceptible root surfaces which become carious. Though the groups are not precisely comparable, Table II-131a lists studies not limited to susceptible root surfaces whilst Table II-131b shows the prevalence when these criteria are applied.

In general, the RCI figures shown in Table II-131b reveal an increase with advancing years. This is what one would expect since older patients have more root surfaces at risk and that have been at risk for some time.

Group	Investigator	No of Patients	% RDF	Age (Yrs)
General Community				
- non-fluoridated	Hazen et al (1972)	500	39	18-82
	Stamm & Banting (1980)	465	36	18+
	Katz et al (1982)	473	42	18-69
	Vehkalahti et al (1983b)	5028	17	30-70+
	Stamm et al (1990)	967	22	18+
- fluoridated	Stamm & Banting (1980)	502	21	18+
	Burt et al (1986)	151	(optimum FL) 23.8	39.8 (mean)
		164	(excess FL) 7.3	43.2 (mean)
	Stamm et al (1990)	967	35	18+
Military Personnel	Sumney et al (1973)	172	44	40-59
	Lohse et al (1977)	281	15	20-70+
Periodontal Patients				
- treated	El-Hadary et al (1975)	220	42	30-60
	Hix & O'Leary (1976)	120	45	40-60
- untreated	Hix & O'Leary (1976)	124	58	40-60
	Ravald & Hamp (1981)	35	87	34-73
Chronically ill, Institutionalised	Sumney et al (1973)	135	49	30-59
	Banting et al (1980)	59	36	65+
	Kiyak et al (1993)	1063	36	72-98
Older Adults, Non-institutionalised	Beck et al (1985)	520	63	65+
	Locker et al (1989)	183	57	50-75+
	Wallace et al (1988)	603	70	60+
	Luan et al (1989)	1744	65	20-80
	Fure & Zickert (1990)	463	89	55-75
	Hellyer et al (1990)	146	88	55+
	Beighton et al (1991c)	146	88	55+
	Douglass et al (1991)	not stated	22	70+
	Fejerskov et al (1991)	90	60	60-80
	Papas et al (1992)	326	33	>middle age
Primitive Tribesmen	Schamschula et al (1972)	22	72	not known
Inuit Elderly	Galan et al (1993)	54	43	60+
Drug Addicts vs Non Addicts Among Female Prisoners	Hecht and Friedman (1949)	151	Addicts 86	34.7 (mean)
		50	Non Addicts 6.3	33.3 (mean)

Table II-131a: The Prevalence of Root Caries and/or restored lesions (% RDF) in a variety of population groups aged 18+ years

Group	Investigator	Age Range	RCI
Non-fluoridated Community	Katz et al (1982)	18-29 yrs 30-39 yrs 40-49 yrs 50-59 yrs 60-69 yrs	1 5 13 22 17
Fluoridated Community	Burt et al (1986)	39.8 (mean) (optimum FL) 43.2 (mean) (excess FL)	1.2
Community Dwelling Older Adults			
- USA	Wallace et al (1988a)	60 + yrs	8.1
- Sweden	Fure & Zickert (1990)	55 yrs 65 yrs 75 yrs	13.8 16.1 21.5
- UK	Hellyer et al (1990)	55 + yrs	17.3
- Norway	Gustavsen et al (1988)	20 + yrs	21.0
- USA	Papas et al (1987)	< 75 yrs 75 + yrs	12.5 14.9
Inuit Elderly	Galan et al (1993)	60 + yrs	19.0
Periodontal Patients	Keltjens et al (1988)	22 + yrs	6.3

Table II-131b: The Prevalence of Root Caries identifying susceptible root surfaces only (RCI) in a variety of groups aged 18 + years

However, the patients in the oldest groups of all show reduced incidences since their root surfaces will have been exposed to the oral environment for some years, so that those which were likely to have become carious will have succumbed some years earlier, whilst those remaining will be less vulnerable for one reason or another. It is perhaps comparable to the well recognised fact that coronal enamel caries is unlikely to be found on smooth accessible tooth surfaces even though they are within the oral environment, other local factors are required. Thus, though all exposed root dentine is 'available' to become carious it cannot be assumed that it is all 'susceptible' to caries.

The prevalence of Primary Root Caries has also been assessed by recording the mean numbers of decayed or filled teeth per person (Table II-131c). Depending on age, these figures show that they range from one to almost ten and that, though the disease per capita is high (40 percent), the prevalence per tooth is low. Some studies suggest that only about 15 percent of all teeth with exposed root dentine exhibit Primary Root Caries, the mean number of teeth affected, per person, being about 2.8 (Katz, 1981; Nyvad and Fejerskov, 1982; Katz et al, 1985).

Though there are wide variations in the published prevalence rates for Primary Root Caries, they do appear to increase with age, with the exceptions referred to above. Under 30 years of age only about one in nine of exposed root surfaces are carious, whilst two in three of such surfaces are carious in patients over the age of 60 years.

Group	Investigator	Age range (Yrs)	Mean No
Non-fluoridated Community	Katz et al (1982)	18-29 30-39 40-49 50-59 60 +	0.2 0.6 1.9 3.0 3.4
National Survey - UK	Todd & Lader (1991)	25-34 35-44 45-54 55-64 65 +	0.3 0.8 1.2 1.9 1.9
National Survey - USA	Miller et al (1987)	25-34 35-44 45-54 55-64 65	0.5 0.7 1.2 2.0 3.2
Military Personnel	Sumney et al (1973)	40-49 50-59	1.0 1.7
Inuit Elderly	Galan et al (1993)	60 +	1.4
Rural Chinese Populations	Luan et al (1989)	50-59 60-69 70 +	1.0 1.8 1.5
Rural Kenyan Population	Manji et al (1989)	45-54 55-65	1.1 1.8
Institutionalised Adults	Sumney et al (1973) Banting et al (1980)	40-49 50-59 65 +	1.4 1.1 9.8
Community Dwelling Older Adults - Canada - USA - Sweden	Locker et al (1989) Wallace et al (1988a) Fure & Zickert (1990)	50-64 65-74 75 + 60-80 + 55	1.9 2.7 4.2 3.1 1.4 3.3 3.6

Table II-131c: Mean Numbers of teeth with root caries and/or fillings or average number of root caries lesions in affected persons by age and population group

Once again the picture is less than clear, because it is difficult to make realistic comparisons of the results obtained from different surveys since the criteria of diagnosis and recording vary. These fundamental difficulties are compounded, since other variables such as age, sex, cultural backgrounds, socio-economic characteristics and sample selection are frequently inadequately defined.

II-132 Incidence. The incidence of Primary Root Caries is defined as the number of new lesions per 100 surfaces of root dentine. The reported figures are summarised in Table II-132.

As with prevalence studies, comparisons are difficult to make in view of the diversity of the populations studied, not least the inclusion in some of younger individuals (Gustafsson et al, 1954; Ravald et al, 1986; Leske and Ripa, 1989; Ravald and Birkhed, 1992). However, the diagnostic criteria employed exhibit less variability, possibly because these studies have been made by a much smaller group of researchers. Nevertheless, variations in the criteria employed are evident with respect to: Location, multiple surfaces; and newly placed restorations. Hand et al (1988a, b) observed that 52 percent of the incremental increase was caused by sound root surfaces becoming carious with the remaining 48 percent becoming restored; but Leske and Ripa (1989) recorded 65 percent of previously sound surfaces becoming restored. Clearly, it is essential that all base-line examinations record abraded non-carious root surfaces separately from non-abraded sound surfaces since it is impossible retrospectively to know whether the condition that indicated the need for a restoration to be placed was caries or abrasion.

Investigator	Population Observed	Duration of Study	% Subjects Developing Root Caries	Attack Rate
				Lesions/100 surfaces at risk
Ravald et al (1986)	Periodontal Patients	4 yrs	62	1.1
Leske & Ripa (1989)	Community-Dwelling Adults 20-65 years	3 yrs	18.6	0.87
Hand et al (1988b)	Community-Dwelling Older Adults in Iowa	3 yrs	43	1.8
Banting et al (1985)	Chronically ill, Older Adults, Institutionalised	2.8 yrs	36	6.3
Hand et al (1988a)	Community-Dwelling Older Adults in Iowa	1.5 yrs	<50	2.6
Wallace et al (1988b)	Community-Dwelling Older Adults in Alabama	1 yr	not stated	1.6
				Lesions/person /period
Gustafsson et al (1954)	Mentally Deficient 37+ yrs, Institutionalised	5 yrs	N/A	0.51
				Mean new root DFS/year
Ravald & Birkhed (1992)	Periodontal Patients	2 yrs	50.5	1.0-1.5

Table II-132: Sample of Incidence Rates of Root Caries in Selected Populations

The studies referred to in Table II-132 suggest that the incidence of Primary Root caries in older individuals is about 2.0, though Banting's study of older, chronically ill and institutionalised patients, records more than three times this figure. This is not surprising since such patients are unlikely to have either the physical ability or the motivation to practice adequate preventive dentistry. The lower than average figures reported by Ravald et al (1986) and Leske and Ripa (1989) can be explained by the inclusion in their studies of younger populations than other researchers. With these exceptions, the caries increments reported are surprisingly similar despite differences in the populations studied and the methodologies employed.

Though studies of healthy ambulant individuals have not been reported, for all those referred to are generally considered to be of populations at high risk of Primary Root Caries, it is likely that their incidence rates are significantly lower. This fact alone indicates the need to design studies of Primary Root Caries with great care, especially any clinical trials, which will require both large sample sizes and adequate statistical power (Banting, 1986). Katz (1985) has suggested that multi-centre trials may be an appropriate approach to overcome such problems. It is, as yet, unclear from the literature just how long it takes for Primary Root Caries to develop to the point of being clinically diagnosable. The exposure of root surfaces to the oral environment undoubtedly occurs insidiously over many years with ever increasing areas being revealed. Banting (1986), therefore, expressed the view that the relationship between the incidence of Primary Root Caries and age would be linear. For this to be the case, the rate of root area exposure would need to be constant,

which is unlikely, and the aetiological factors of diet and plaque control would also need to be constant. To determine, with any degree of accuracy, the rate of root surface exposure, as distinct from the linear recession of the gingival attachment, would be remarkably difficult in view of the range of Size and shapes found in the human dentition. Furthermore, an assumption that all exposed root dentine is equally susceptible to caries is unlikely to be true, for once it is exposed it is likely that its susceptibility will vary with time, and dentine which has resisted caries attack for some time may well continue to be resistant for an indefinite period (Banting, 1986).

II-200 Aetiology

II-210 Root Exposure

By definition, Primary Root Caries is that which is initiated in the dentine of the roots of teeth as a consequence of bacterial activity. The aetiology of coronal caries has been more extensively investigated than that of Primary Root Caries and it has generally been assumed that they are comparable. Newbrun (1986) stated that 'Root caries is undoubtedly of microbial origin' and Jordan (1986) accepted this but considered that interactions between oral bacteria and other factors involved in the development of Primary Root Caries may be broader in scope and more difficult to interpret than those which result in coronal caries. On the assumption that this is correct, root dentine (cementum) needs to be accessible to the oral environment, ie there must have been some loss of gingival attachment to expose root surfaces. Such exposure is, clearly a predisposing factor for without it, by definition, Primary Root Caries is impossible, but it cannot logically be described as a true aetiological factor.

It will come as no surprise that root caries is observed predominantly in age groups where loss of periodontal attachment and subsequent gingival recession has occurred (Baelum and Fejerskov, 1986; Baelum et al, 1988). It may be expected that the greater the area of root dentine exposed to the oral environment, the greater will be the incidence of Primary Root Caries. This has also been reported by McDermott et al (1991) who identified loss of attachment of 3 mm or more as of significance to the Primary Root Caries that may develop; by Galan et al (1993); and by Guggenheim and Lutz (1985) who induced periodontal breakdown in rats

and found a corresponding increase in Primary Root Caries experience. However, it would appear to be quite illogical to conclude that those of advanced years are more susceptible to Primary Root Caries as a consequence of their age (Ravald and Hamp, 1981) when perhaps the major factor is simply that the elderly are likely to have more root dentine exposed than do others. Velden (1984) noted that ageing is accompanied by a variety of periodontal changes, notably that the degree of periodontal breakdown increases with age. It may also be inappropriate to consider the degree of recession as a measure of risk of root caries in a population as borne out by Luan et al (1989). Here the population was observed to have extensive gingival recession with age, but little root caries, with only 10 percent of exposed root surfaces or surfaces 'at risk' showing root caries. Gustavsen et al (1988) could not entirely relate the increase in RCI with the increase of gingival recession with age.

The paper by Hix and O'Leary (1976) reports results which do not show that increased root surface exposure will tend to reveal more Primary Root Caries. They studied patients who had undergone periodontal surgery resulting in increased exposure of root dentine but found no corresponding increase in caries incidence. This was probably due to two main factors; firstly, any patient who was prescribed periodontal surgery would undoubtedly have achieved a high level of plaque control before it was undertaken, and if this was not the case, their plaque control after surgery would undoubtedly be beyond reproach; and secondly, incipient or early caries would have been identified and/or been removed by 'Root planning', an essential component of surgical periodontology.

II-220 Diet

II-221 Readily Fermented Carbohydrates and pH. As long ago as 1954, Gustaffson et al, in their Vipeholm study reported a higher incidence of Primary Root Caries in older subjects following a regime of frequent intakes of refined carbohydrates than in control groups. Indeed, these lesions accounted for 25 percent of all lesions developing in this group. Similar conclusions were drawn by Hix and O'Leary (1976) who analysed the diet histories of individuals for the frequency of ingestion of readily fermentable carbohydrates. All the individuals studied had significant areas of root dentine exposed as a consequence of either treated or untreated periodontal disease. Individuals with the most Primary Root Caries also gave a pattern of more frequent ingestion of these foods, though the amounts or types of carbohydrates were not specified.

The different experiences of coronal and Primary Root Caries observed in anthropological studies has been referred to (Miles, 1969 and Moore and Corbett, 1973). Schamschula et al (1974) also noted that Primary Root Caries was more predominant than coronal caries amongst the natives of New Guinea whose diet consisted of high proportions of the less refined sugars found in fruit and sweet potatoes. Presumably, the intake of less refined carbohydrate enabled teeth to be conserved intact as long as enamel only was at risk of caries but, once poor oral hygiene resulted in a loss of periodontal attachment, the exposed root dentine, being more susceptible to caries than enamel, succumbed. The damaging consequences of a refined carbohydrate diet through Primary Root Caries was shown by Tavares et al (1991), who found that whilst diabetics have a high incidence of periodontal disease with the consequent exposure of root

dentine, their level of Primary Root Caries was lower than a non-diabetic control group. They inferred that this was probably due to the strictly controlled low intake of refined carbohydrate by the diabetics.

The connection between refined carbohydrate intake and the pH of dental plaque was first demonstrated by Stephan (1944) in a classic experiment which demonstrated its rapid fall after a sugared drink. Abelson and Mandel (1990) repeating a similar exercise on just twelve subjects claimed that the pH readings of plaque on the surfaces of the roots of teeth were consistently lower than from that on enamel. Hoppenbrouwers et al (1986, 1987) showed that the root dentine of extracted teeth may decalcify at the near neutral pH of 6.7, compared with enamel which requires a pH of 5.5. However, these root surfaces would not have been exposed to the oral environment and potentially undergone any changes that this might have brought about. This would include the remineralisation of carious lesions bathed in saliva observed by Kidd and Joyston-Bechal (1987). In 1989, Surmont and Martens reported similar findings to those of Hoppenbrouwers et al (1987), ie that root dentine demineralises at pH 6.7 compared with enamel at 5.4.

The role of saliva in the development of Primary Root Caries, notably the adverse consequences of an inadequate flow, have been observed clinically for many years. This has been assumed to be due to saliva's ability to influence the cariogenicity of dental plaque; to its buffering properties which tend to keep plaque at a near neutral pH; to its potential ability to remineralise carious dentine; and to certain bacteriostatic qualities. Changes in salivary flow have, therefore, been of interest in Primary Root Caries research. Patients who had received radiation therapy to the head

and neck with a side-effect of reduced salivary flow, were found to have significantly higher incidences of both coronal and Primary Root Caries (Katz, 1982) and others on xerostomic drug therapies also have been shown to have increased Primary Root Caries (Streckfus et al, 1985, and Kitamura et al, 1986) and coronal caries as well (Papas et al, 1993). However, Beighton et al (1991c) reported the opposite results from their studies.

II-222 Fluoride. The beneficial consequences of appropriate levels of both systemic and topically-applied fluoride on coronal enamel caries have been the subjects of very extensive research over the past thirty years or more, but any such effects on Primary Root Caries have been infrequently investigated. However, Burt et al (1986), Stamm and Banting (1980) and Stamm et al (1990) have reported that, amongst the populations living in areas served with naturally fluoridated water supplies, lower than average levels of Primary Root Caries were diagnosed. Jensen and Kohout (1988) and Ögaard et al (1990) also claimed that reduced levels of Primary Root Caries have been observed when the topical application of fluoride was prescribed.

II-230 Removable Dentures

The wearing of removable partial dentures clearly change the ecology of the oral cavity resulting in increased plaque accumulation (Carlsson et al, 1970; Brill et al, 1977 and Stipho et al, 1978); Carlsson et al (1970) also demonstrated that the intra oral pH is lowered. An expected consequence of such changes: higher than average levels of Primary Root

Caries were recorded by Wright et al (1992). Similarly, in a random sample of 1019 elderly people living in residential homes, higher levels of both coronal and Primary Root Caries were recorded for those wearing removable partial dentures when compared with those who did not wear such appliances (Drake and Beck, 1993). This was especially so in abutment teeth. Finally, Grippo and Masi (1991) have hypothesised that tooth flexure, stress concentration, stress corrosion, and piezo-electric currents may play their parts in the aetiology of Primary Root Caries. However, these suggestions are made only in the light of a very preliminary study.

II-240 Micro-organisms

Results of early animal experiments led to the conclusion that root caries was associated with a different microflora to that of coronal caries (Keyes, 1969). Whilst hamster and gnotobiotic rats infected with specific strains of Lactobacilli or Streptococci developed enamel caries (Fitzgerald and Keyes, 1960; Fitzgerald, Jordan and Archard, 1966), experimental animals infected with various strains of Actinomyces in contrast developed root caries but no enamel caries (Jordan and Keyes, 1964; Socransky, Hubersak and Propas, 1970).

Enamel caries is now firmly believed to be associated with specific organisms: the Lactobacilli and the Mutans streptococci notably *Streptococcus mutans* and *Streptococcus sobrinus* (Emilson and Krasse, 1985) and, though Newbrun (1986) stated that carious dentine from Primary Root Caries is undoubtedly of microbial origin, any precise causative agent

is more difficult to identify. Sampling to ascertain causative microflora can clearly be of three kinds: the saliva which provides the overall environment of exposed root surfaces; and plaque which lies on the surface of such roots; or Primary Root Caries itself. The fundamental problem lies in the fact that the periodontal attachment needs to be destroyed for root surfaces to become exposed and this is usually the result of periodontal diseases. These surfaces; the soft tissues of the gingiva; and the gingival crevice or pocket are all very closely located to each other, so any bacteriological sample could identify organisms associated with either periodontal disease, Primary Root Caries, or both. Nevertheless, Bowden (1990) states that "Given careful characterisation of the flora and the state of the lesions, it is likely that microbiology will be useful in diagnosing the state of a root surface lesion and in monitoring the use of fluoride and antibacterials in control of the progression of root surface caries."

Though published work has referred to numerous different micro-organisms as being associated with the development of Primary Root Caries, three groups would seem to have come under particular scrutiny: the Mutans streptococci; the Lactobacilli; and the Gram-positive pleomorphic rods (GPPR); whilst some authors have also suggested that certain Yeasts have a role (Beighton et al, 1991c). Some other authors have looked at Yeasts but found no significant association with root caries (Billings et al, 1985; Brown et al, 1986; Hellyer et al, 1990: Collier et al, 1992). Bowden (1990) published a helpful list of organisms which various researchers found to be associated with Primary Root Caries. This included the following to which the results of more recent studies have been added:

Streptococcus mutans:	Syed et al	(1975)
	Ellen et al	(1985a, b)
	Brown et al	(1986)
	Keltjens et al	(1987a)
	Emilson et al	(1988)
	van Houte et al	(1990)
	Bowden et al	(1990)
	Sansone et al	(1993)
Lactobacillus:	Ellen et al	(1985a, b)
	Brown et al	(1986)
	Keltjens et al	(1987a)
	Emilson et al	(1988)
	Bowden et al	(1990)
	Emilson et al	(1993)
Actinomyces viscosus Serovar 2:	Jordan and Hammond	(1972)
	Sumney and Jordan	(1974)
	Syed et al	(1975)
	Ellen et al	(1985a, b)
	Brown et al	(1986)
	Bowden et al	(1990)
Actinomyces naeslundii:	Jordan and Hammond	(1972)
	Sumney and Jordan	(1974)
	Syed et al	(1975)
	Ellen et al	(1985a, b)
	van Houte et al	(1990)
'Intermediate' Actinomyces:	Jordan and Hammond	(1972)
	Syed et al	(1975)
	Ellen et al	(1985a, b)
	Brown et al	(1986)
Actinomyces viscosus/naeslundii:	Keltjens et al	(1987a)
	Emilson et al	(1988)
	Emilson et al	(1993)

However, Bowden (1990) stated that heterogenous species or cariogenic biotypes may be hidden within what is currently named *A viscosus*. Rearrangements in classification would cast doubt on previous clinical microbiological studies which would render the previously used identification schemes outdated. There may be metabolically different *A viscosus* biotypes colonising Primary Root Caries lesions as compared with those in plaque overlying sound root surfaces, and it is possible that lesions select for metabolic adaptations of the *A viscosus* strains established on them. Bowden et al (1990) suggest that careful taxonomic and physiological classifications of *Actinomyces* spp could reveal a role for specific strains in the aetiology of root caries. The importance of specific .*Actinomyces* spp in root caries has to be viewed with caution in the light of the changing taxonomy of this genus in particular (Johnson et al, 1990). Other genera which have been associated with root caries include Veillonella, Bifidobacterium, Rothia, Capnocytophaga, Bacteriodes, Arachnia, Selenomonas and Fusobacterium (Bowden, 1990)

II-241 Microbiology of Saliva. The number of Mutans streptococci in dental plaque can be reflected in the numbers grown from saliva (Togelius et al, 1984). On this premise, several studies have found associations between caries experience and increment and salivary levels of Mutans streptococci, Lactobacilli, and Yeasts, as reviewed by Hamada and Slade (1980) and Emilson and Krasse (1985). Positive associations have tended to be from industrialised countries where there is a relatively high caries activity (Littleton et al, 1970; Klock and Krasse, 1977; Newbrun et al, 1984). Where there is less caries activity, weaker associations are found. The low level of disease activity influences the positive predictive

value and should be borne in mind when making conclusions from research involving saliva sampling. The collection of samples of saliva is relatively easy and, since a salivary sample represents the overall environment of exposed root surfaces it is logical to study its microbiology and relate this to the development of Primary Root Caries. Ravald et al (1986) state that the most prevalent risk factors for root caries in an eight year study were for those patients with high Lactobacilli and Mutans streptococci salivary counts as well as dietary factors.

Van Houte et al (1990) carried this type of investigation further by culturing saliva samples from 33 percent of 273 individuals which also included dental plaque samples from tooth surfaces for both Mutans streptococci and Lactobacilli, and relating the results to clinical observations of the Primary Root Caries experienced. They found that patients with clinically diagnosable Primary Root Caries had significantly higher saliva concentrations of Mutans streptococci and higher prevalence and proportions of Mutans streptococci in plaque than those without it, whether or not restorations were present on the root surfaces, but that these counts did not rise in relation to the numbers of lesions found. On the other hand the prevalence and proportions of Lactobacilli in plaque associated with sound and carious root surfaces were very low and did not appear to be significantly higher where Primary Root Caries was diagnosed.

Hunt et al (1992) carried out comparable investigations on 448 Black and 362 white patients and were able to confirm higher levels of both *S mutans* and Lactobacilli in the saliva of those with higher than average number of untreated Primary Root Caries lesions. However, their total Primary Root Caries experience was no higher. Of potential significance to

the management of Primary Root Caries are the findings of Emilson et al (1993) who reported that a regime of oral prophylaxis and the application of a fluoride-containing varnish at three monthly intervals over a twelve-month period failed to alter the levels of both Lactobacilli and Streptococcus mutans in the saliva of 15 patients with some active Primary Root Caries. Furthermore, whilst 53 sites originally diagnosed as being active lesions were later considered to be inactive an almost equal number, 55, were diagnosed as changing from inactive to active.

The studies which associate salivary levels of these two organisms with the levels of Primary Root Caries seem to be questioned by the work of Beighton et al (1991c) who examined 146 patients all over 55 years and with a minimum of 12 standing teeth. They found a stronger relationship between the presence of Yeasts in saliva and the presence of Primary Root Caries than the presence of Mutans streptococci and Lactobacilli. However, they do not claim a causative role for Yeasts nor that the other organisms do not have an aetiological role in the initiation and progression of these lesions. They also observed that, although the levels of all three genera of organisms were raised significantly in the saliva of those patients wearing dentures, the relationship of the organisms to root caries was the same.

Many other authors have also related salivary *S mutans* and Lactobacilli with root caries (Ravald and Hamp, 1981; Ellen et al, 1985a, b; Ravald et al, 1986; Fure et al, 1987; Keltjens et al, 1987a, b, 1988; van Houte et al, 1990).

Dental caries is a relatively slowly progressing pathological condition; the saliva can be considered to be only the general background environment of exposed root dentine; whilst the bacteriological flora at its surface is likely to be more relevant than that of saliva.

II-242 Microbiology of Plaque on Root Dentine Surfaces. Syed et al (1975) collected 21 plaque samples mixed with some carious debris from the surfaces of Primary Root Caries in the mouths of 15 patients. It was stated that these patients had undergone periodontal treatment, some surgical, but no attempt was made to assess the activity levels of the lesions identified. *S mutans* was identified in 13 of the 21 samples, which also contained *A viscosus* (30 percent and 47 percent of the total colony forming units respectively); whilst the remaining 8 samples were free of *S mutans* but consisted of *S sanguis* (48 percent) and *A viscosus* (41 percent) of all cultivable bacteria. Thus, *A viscosus* accounted for over 40 percent of the viable count of all the plaque samples studied and was by far the most dominant of the *Actinomyces spp* identified (*A viscosus*, 42; *A naeslundii*, 5; and another 5 could not be typed), but in one group *S mutans* was prevalent with 30 percent of the colony forming units, to be replaced by *S sanguis* (48 percent) in the second group. These authors concluded that whilst *A naeslundii* had been reported to be involved in root caries and periodontal disease in animal studies (Jordan and Hammond, 1972; Socransky et al 1970), their own study could not confirm it as a predominant organism from root surface plaque. They mooted the possibility that it may be confined to softened dentine rather than plaque overlying root caries lesions.

In a 1977 study by Hill et al, plaque samples collected from all accessible coronal and radicular surfaces, excluding gingival crevices revealed a dominance of streptococci (26 percent) and *Actinomyces spp* (33 percent) of colony forming units. Of the total count of those streptococci 68 percent were anaerobic and 70 percent aerobic but the investigation concentrated on typing the *Actinomyces spp*: 42 percent were *Actinomyces odontolyticus*; 28 percent *Actinomyces israelii*; 14 percent *A viscosus*; and 9 percent *A naeslundii*. The relatively lower preponderance of *A viscosus* (14 percent) compared with the 41 percent reported by Syed et al (1975) could be due to the fact that the plaque samples studied in the latter were collected from the surfaces of lesions only whilst the former were from all surfaces of teeth.

Ellen et al (1985a) also linked *S mutans* in dental plaque with Primary Root Caries but also involved Lactobacilli, finding that more than 80 percent of root dentine surfaces which were diagnosed as non-carious at the first visit but from which plaque was retrieved which contained these organisms, were diagnosed as being carious at later visits. On the other hand virtually no surface related to plaque which was free of these organisms became carious. Brown et al (1986) also identified *S mutans* in the plaque overlying early and advanced Primary Root Caries but reported that they constituted 80 percent of the total streptococcal count from early lesions as against less than 45 percent from advanced lesions, inferring different plaque characteristics for different stages of lesions.

The presence of *S mutans*; *S mutans* and *Lactobacilli*; and/or *S mutans* and *Actinomyces spp*, in the plaque overlying Primary Root Caries at various stages has been reported by many other authors, notably

Billings et al (1985), Keltjens et al (1987a, b); Nyvad and Kilian (1987); Fure et al (1987); Emilson et al (1988, 1993); MacEntee et al (1990); van Houte et al (1990); Bowden et al (1990) and Sansone et al (1993). Whilst the plaque overlying most root surfaces is likely to be colonised by one or more of these types of organisms, many of these researchers consider the numbers to be higher when the surface is carious and that the dentine is more likely to be affected when more than one type is present in significant numbers.

II-243 Microbiology of Carious Dentine. Clearly the microbiology of saliva and of plaque from the surfaces of the exposed roots of teeth can only be seen as potential predisposing factors in the development of Primary Root Caries and, therefore, as possible predictors of future disease. The microbiological characteristics of affected tissue may be quite different. The problems associated with obtaining dentine samples uncontaminated with plaque will be discussed when considering the methods employed in the current study, but it is clear that, in some studies, samples were perhaps less pure than they were assumed to be (Syed et al, 1975; Brown et al, 1986; Bowden et al, 1990). Carious dentine itself has not been studied as extensively as either plaque or saliva, possibly due to the fact that tissue needs to be removed and such research would carry with it treatment implications for which the investigators, often microbiologists, could not accept responsibilities; nor could such information be considered of value as a predictor, for the disease was already established.

Jordan and Hammond (1972) avoided clinical management implications by sampling root caries lesions in extracted human teeth.

Strains of *Rothia dentocariosa, A viscosus, A naeslundii, A odontolyticus* and *Actinomyces erisonii* were identified and a third of *Actinomyces spp* isolates could not be classified. Gram-stained cemental lesions when sectioned revealed Actinomyces-like bacteria invading the length of the dentinal tubules and laterally between tubules. This pattern had already been noted by other workers on animal models involving *A viscosus* (Keyes and Jordan, 1964; Jordan and Keyes, 1964). Sumney and Jordan (1974) made great efforts to obtain discrete samples of both the surfaces of lesions and the advancing fronts of lesions proximating the underlying sound dentine, again in extracted teeth. In surface dentine *S mutans* was found to be a significant component of the bacterial flora along with fifteen strains typical of the genus *Actinomyces* (*A viscosus* (6); *A naeslundii* (2); *Actinomyces odontolyticus* (3); and (4) unidentified species). Carious dentine nearer to the advancing front of the lesion revealed the presence of aerobic diptheroids (Arthrobacter) which contrasted with studies showing the predominance of Lactobacilli at the advancing front of coronal dentine caries. Since the acidogenic potential of Arthrobacter species is low it is surmised that they might be responsible for alterations to the organic matrix of the dentine following its decalcification. The authors claimed that their work showed that unique bacterial flora are present in carious lesions arising on different surfaces of the teeth and especially that the flora in Primary Root Caries is different to that in other Locations.

Nyvad and Fejerskov (1987a, b) found Gram-positive bacteria with thick cell walls in the carious dentine close to the surface of roots and between collagen fibres which they thought were *Actinomyces spp.* The samples of dentine were obtained from the surfaces of tooth fragments

attached to intra-oral appliances worn by dental students for up to 48 hours during which period they suspended all oral hygiene measures and the samples were studied by light, transmission electron, and scanning electron microscopy. Though these organisms were dominant within the dentine the overlying plaque contained mostly coccoid forms. Comparable results were also reported by Nyvad and Kilian (1987) using conventional microbiological techniques.

In an early attempt to identify where micro-organisms are located in Primary Root Caries, Sumney and Jordan (1974) examined sections of carious dentine. Gram-positive organisms, which formed a dense layer on the surfaces of roots, were also found in the carious dentine both within and between the dentinal tubules, but near the advancing fronts of lesions they were limited to the tubules. Presumably, this was simply a manifestation of their ability to progress down open non-calcified tubules even before the dentine matrix around them was decalcified, after which their entry into the latter can occur. Five years earlier, Furseth and Johansen (1968), then Westbrook et al (1974), using electron microscopy claimed that sub-surface decalcification occurred as an early manifestation of Primary Root Caries, as is known to be the case with enamel caries, but Frank et al (1989) were unable to confirm this. These latter authors studied superficial Primary Root Caries, also using electron microscopy and described how great numbers of predominantly Gram-positive organisms, arranged with coccoid bacteria surrounding a sheaf of filamentous organisms, were present in the plaque on root surfaces. Bacterial infiltration of cementum mainly occurred down incremental lines and between calcified bundles but no demineralisation gradients could be detected in that carious tissue.

These narrow channels first became filled with single rows of Gram-positive bacteria then a simultaneous destruction of both the inorganic hydroxyapatite component and the collagen matrix of the cementum resulted in a widening of these channels. In contrast, caries of the root dentine did involve a demineralisation gradient, but again bacterial invasion was initially down naturally-occurring channels, in this case the dentinal tubules which become packed with Gram-positive organisms and were enlarged as a consequence of the destruction of the nearest dentine matrix: the well calcified peritubular matrix. As this destruction of peritubular matrix continued a diffused decalcification of the intertubular matrix proceeds revealing the large, typically cross banded, collagen fibrils of that tissue. Eventually, the destruction of the intertubular matrix results in the confluence of even larger tubules packed with micro-organisms and the total destruction of the root dentine is evident.

Nyvad and Fejerskov (1987b) carried out similar studies to relate the presence and Locations of micro-organisms in Primary Root Caries but included comparisons between what were judged clinically to be active or inactive lesions. Lesions which were described as yellowish and Soft were designated as active, most of which revealed a hypermineralised surface layer overlying the main demineralised body lying beneath. Also at the advancing front of the lesions was a narrow zone of demineralisation whilst within the main body of the lesion zones described as sclerotic, presumably meaning more radio-opaque than surrounding dentine, were evident. In contrast with this description of active caries, lesions which were judged clinically to be Hard and designated as inactive, displayed a uniform distribution of mineral content throughout, apart from a few small areas of

sub-surface demineralisation. Between these two extreme types of lesion were those which were clinically diagnosed as being dark, with a Leathery Texture but designated as active. These displayed a wide range of histological appearances, some revealing a gradual increase in radiopacity from their body to their pulpal front. The authors postulated that any change from an active to an inactive lesion was probably associated with the loss of its outer, softer layers, together with an uptake of minerals from oral fluids and that the absence of a well mineralised surface layer indicates an active lesion.

The same researchers (Nyvad and Fejerskov, 1990) induced Primary Root Caries in samples placed in the flanges of dentures which had all the clinical characteristics of active lesions: Yellow to Light Tan in Colour and with a Soft Texture. On micro-radiographic examination, the cementum was intersected by numerous fine radiolucent channels perpendicular to the root surface which in some zones coalesced to produce localised micro-cavities, but the mineral distribution at the surface of any lesion varied considerably within short distances. The demineralisation of cementum displayed a gradient from maximum at the root surface reducing towards the dentine surface, they did not find the dense mineralisation described by others. Bacterial invasion occurred along the borders between bundles of relatively well-mineralised extrinsic collagen fibres in which the characteristic cross-banding remained intact. The pattern of bacterial invasion was influenced by the incremental lines and the cemento-dentinal junction. These bacteria were nearly all Gram-positive. It was suggested that the level of activity in a carious lesion in cementum could be determined by examination of the ultrastructure.

Schüpbach et al (1989) examined micro-radiographically early Primary Root Caries, mainly of cementum, in 24 extracted non-restored teeth from eight subjects aged 56-77 years. Cementum surfaces were found to be covered by thick layers of micro-organisms, from which strips of enhanced radiolucency often extended to the dentine and into its superficial layers, when the surface of the cementum was irregular; alternatively the surface contour remained intact, though depressed, and an even demineralisation of both cementum and dentine was revealed, though the cementum retained a higher mineral content than the dentine. Alternatively, one hypermineralised surface layer was evident overlying less radio-opaque cementum and a second overlying the decalcified dentine. As a lesion developed, cementum fractures along the incremental lines were observed which sequentially destroyed the most superficial layers. Loss of cementum was also observed immediately adjacent to any micro-organisms. A comparable step-wise demineralisation of dentine was also observed: first a uniform partial demineralisation was seen with a convex front, the gradient being decreasing demineralisation from the root surface. As the lesions developed, a 'halo-like', virtually completely demineralised zone was noted extending from the root surface into the partially demineralised zone. The narrow layer of superficial dentine devoid of tubules then developed small clefts in the carious tissue along which micro-organisms made their first penetration of dentine.

More advanced Primary Root Caries have been examined by Schüpbach et al (1990a, b). Again, micro-organisms were identified in zones of bacterial penetration, travelling first along the dentinal tubules then, as the peritubular and intertubular matrices were decalcified, they

moved into them, resulting in a massive infection of the whole tissue. This is a gradual process, so that zones of demineralisation could be identified devoid of micro-organisms in which there was virtual total destruction of most peritubular matrix but only partial demineralisation of intertubular matrix, though some large crystals of hydroxyapatite from the hypercalcified part of the peritubular matrix may survive. Zones of so-called tubular sclerosis were also evident adjacent to zones of demineralisation where the hyper-calcification associated with many parts of the peritubular matrix had obliterated the dentinal tubules either partially or totally . Evidence of radiolucency in the hypercalcified peritubular matrix around such tubule deposits suggest that some decalcification occurred before these deposits were laid down. A consequence of the bacterial penetration of carious dentine is its destruction. The outermost layers became fragmented with large clefts developing radially and, in these, large numbers of micro-organisms could be identified, generally embedded in the dentine matrix but dentine morphology was identifiable. The clefts frequently extended into the dentine with sclerosed tubules and eventually their highly mineralised centres were destroyed.

Finally, lesions in extracted teeth diagnosed as 'arrested' have been studied by Schüpbach et al (1992). It was found that in such lesions intertubular matrix was 'fully' mineralised throughout the carious dentine and tubules were 'sclerosed' in the surface layers. The sclerosis of the dentinal tubules revealed a number of different appearances many including remnants of the processes of the odontoblasts or of the collagen matrix, others were filled with the 'ghosts' of micro-organisms, whilst the types of crystals observed also varied. The authors suggested that an

active lesion becomes arrested due to the development within the dentine of a layer which prevents diffusion from the pulp; a compact, highly mineralised, superficial layer which prevents diffusion from the oral cavity; and a third barrier of highly mineralised tissue, thus enclosing the lesion from the root surface to the pulp. By such means, micro-organisms within the carious dentine become starved and the decalcification of dentine arising from the products of micro-organisms in plaque lying on the root surface is inhibited. These observations and conclusions are, of course, precisely in line with and support the basic principles that have been the foundation of the management of all types of caries by clinicians for generations.

II-244 Conclusions. Though a significant variety of organisms have been implicated in the development of Primary Root Caries, either from their presence in saliva; in dental plaque on the surfaces of roots; or within the carious dentine itself, it would seem that the general findings tend to implicate the Mutans streptococci; the Lactobacilli and the Gram-positive pleomorphic rods (*Actinomyces spp*); whilst some role, perhaps secondary, seems to exist for the Yeasts. Its aetiology would then be comparable to that of coronal caries but, not surprisingly, root dentine is more susceptible to the carious process than is enamel. Some authors appear to confuse true aetiological factors with predisposing factors, which is unfortunate. Aetiological factors such as those described are of significance to the prevention and management of Primary Root Caries and, whilst some can forewarn clinicians and epidemiologists of the likelihood of the disease being present, they do not help in its precise diagnosis.

Valid comparisons of studies are made difficult because of the diverse sampling techniques used, which as Nyvad and Kilian (1990a) pointed out takes no account of the possible variations in the composition of the microflora across each lesion, and that nearly all studies use selective media for enumeration of target organisms, which may limit the true picture of the microflora.

There is a lack of definition of the types of lesions under investigation in the studies, which fails to take into consideration the dynamic nature of the disease process.

The histological evidence of the patterns of microbial attack and of the changes in the mineralisation of cementum and dentine are of interest and relevance to this study, for the first is relevant to microbiological studies and the second to the Texture of lesions, one of the most important clinical signs of attack. Many of the characteristics described are understandable with an appreciation of the fundamental structural characteristics of dentine and its ability to respond to microbiological attack. Dentinal tubules at their largest diameter adjacent to the pulp are 4 to 5 microns across and although this is reduced by the deposition of peritubular (intratubular) matrix, the greater part of sound dentine is traversed by tubules much greater in diameter than the micro-organisms which give rise to the disease. Once the tubule system is breached by the destruction and cracking of the thin surface layer of dentine, wide open tubules can rapidly become colonised by micro-organisms. From that point onwards, the carious attack can be made both on a broad front from the surface of the root and from within the tubules, as well as from new centres around the initial site of attack. It could, therefore, be predicted that the advancing

front of any lesion will be convex and this is what both clinical experience and research findings confirm.

The reaction of the pulp-dentine complex to carious attack is the stimulation of both the deposition of organic matrix within the dentinal tubules and its high mineralisation and an increased mineralisation of intertubular matrix. Given the fact that dentine caries is a relatively slow pathological process occurring in the environment of the mouth, which can be favourable for the deposition of mineral salts, it is not surprising that surface layers of carious root dentine can be shown to be more heavily mineralised than the carious dentine beneath, whilst the pulp-dentine response can complete the encapsulation of the caries by the deposition of mineralised matrix on all other aspects. It is logical, to avoid destroying such an encapsulation, when the preferred management strategy might be of a non-operative kind and any sharp instrument such as a probe would undoubtedly bring this about, as Kidd (1990) suggests.

II-250 Risk Factors, Risk Indicators and Predictors of Primary Root Caries

An ability to identify circumstances which predict the likelihood of Primary Root Caries developing is clearly of advantage if steps are to be taken to prevent the disease developing. Prevention of any disease rather than the management of established disease has humane, financial, time, surgical and technical advantages which cannot be denied.

Social, behavioural, medical and oral factors which are associated with the incidence of Primary Root Caries, based on both clinical and laboratory data, have been termed 'Risk Factors' (Beck, 1990), when

relationships are confirmed by longitudinal studies (Joshi et al, 1993). If such confirmation has not been obtained such potential risk factors identified in cross-sectional studies are termed 'Risk Indicators'. The use of multivariate analysis of such Risk Factors and Risk Indicators of Primary Root Caries enables a 'Risk Prediction' to be determined.

Analysing prevalence studies of Primary Root Caries, Beck (1990) identified age as the single most positively associated prominent Risk Indicator; the presence of coronal caries; the fluoridation of the water supply; the degree of loss of gingival attachment and educational levels were also positively associated, but to a lesser extent whilst the numbers of retained teeth were negatively associated.

It has been stated that the increase in the proportion of older people in the population and the increase in the retention of teeth will mean more teeth are at risk of root caries (Vehkalahti, 1987c; Reinhardt and Douglass, 1989). However, though the 1988 UK Dental Health Survey showed that, due to an increased retention of teeth, the pool of teeth at risk had been increased, it was difficult to assess the extent to which this risk would be translated into overt disease (Downer, 1991). Contrary to such an hypothesis, some data suggest that the more teeth that are retained, the lower the Primary Root Caries Risk Factor (Kitamura et al, 1986; Beck et al, 1986, 1988; Beck 1990; Fure and Zickert, 1990; Beighton et al, 1991c; Graves et al, 1992), whilst the inter-dependence of such Risk Factors as the numbers of teeth and age has been demonstrated in the data from the Finnish National Survey (Vehkalahti, 1987c). A substantially lower prevalence of Primary Root Caries was found amongst individuals having more than 21 teeth than amongst those with fewer teeth, but this was only

true when all age groups were assessed together. When the data from various age-groups were analysed separately, this was found not to be the case, but simply that younger patients had more teeth than older ones.

Fejerskov and Nyvad (1992) have suggested that it was logical that the incidence of Primary Root Caries will increase with age, as shown in many studies (Kirkegaard et al, 1985; Miller et al, 1987; Gustavsen et al, 1988; Salonen et al, 1989) and that the age association could legitimately be made as long as it is a simple reflection of more root surfaces being exposed with increasing age. Baelum's Danish study (Fejerskov and Nyvad, 1992) analysed the data from two groups: the 60-70 years and over 70 years of age, by plotting the percent of individuals with Primary Root Caries in each group against the number of teeth at risk. When the numbers of teeth were taken into account, the age dependent increase in prevalence almost disappeared, indicating that, the more teeth that were retained in to old age, the less was the caries prevalence. Beck (1990) also noted that patients who reached 65 years old with many of their own teeth had less Primary Root Caries lesions, whereas those who had nine or fewer teeth by this age were experiencing new lesions. It has been well established that teeth are lost up to the age of about 60 years mainly as a direct result of the caries process (Ainamo et al, 1984; Baelum and Fejerskov, 1986; Bouma et al, 1987; Baelum et al, 1988). Fejerskov and Nyvad (1992) have, therefore, suggested that the more teeth that are retained, the more likely it is that the individual has experienced a low caries progression rate. The risk of developing Primary Root Caries is clearly associated with an individual's past coronal caries experience (Burt et al, 1986; Vehkalati, 1987c; De Paola et al, 1989b; Locker et al, 1989)

and has been positively correlated with baseline Primary Root Caries scores (Ravald et al, 1986; Leske and Ripa, 1989a).

Joshi et al (1993) considered that the available data to assess incidence and associated Risk Factors on Primary Root Caries are sparse and that few studies have been subjected to multivariate analysis. They have called for further investigations and have instituted a two year project involving the clinical assessment of the incidence of Primary Root Caries in 130 middle-aged and elderly adults, and are relating the results to a range of associated Risk Factors using multivariate logistic regression analysis. To date they have identified: past Primary Root Caries experience; high plaque scores; and high numbers of teeth (22 or more) to be positively associated with new Primary Root Caries ($P < 0.05$). Clearly, this is the opposite trend with reference to the numbers of teeth than that reported by the authors stated above. The subjects in the Joshi et al (1993) study regularly attended their dentist; were well educated, had high incomes; and only 18 percent of Primary Root Caries was untreated. These characteristics were in contrast to the subjects of the study by Hand et al (1988a) who, for example, had 52 percent of Primary Root Caries lesions unrestored. Such differing data highlight the fact, noted by other researchers, that tooth loss occurs, not only as a consequence of disease *per se*, but also from a patient's demands (eg extraction rather than restoration), and from dentists' perceptions of need which are governed by their treatment rationale (Hand et al, 1991).

MacEntee et al (1993) have defined three steps that need to be taken to identify variables contributing to risk of a disease:

- social and clinical variables that are potential risks are identified;

- associations between these and the disease are measured using bivariate analysis; and

- multivariate models are constructed and analysed to highlight those risks having the most direct impact on the disease.

Using this method, these researchers identified past caries experience; poor oral hygiene; frequent sugar consumption; high salivary Lactobacilli counts; and residence in long-term care facilities, as the most significant variables contributing to a risk of developing caries, including Primary Root Caries. These results supported the contention of Locker et al (1989) that oral health variables are probably better predictors of caries, including Primary Root Caries, than socio-demographic and general health variables or access to dental care; an opinion disputed by Beck et al (1986). The importance of intra-oral risk factors has also been supported through multivariate analysis by Graves et al (1989), Beighton et al (1991c), Ravald and Birkhed (1991 and 1992) and Sheinin et al (1992), notably with respect to past Primary Root Caries experience; visible plaque; and salivary levels of candida albicans, which were identified as the best predictors. Nevertheless, Galan and Lynch (1993a), in reviewing the many studies investigating the significance of non-oral factors in the prediction of Primary Root Caries, supported the importance of medical, mental, behavioural and social conditions suggested by Beck (1990).

II-300 Management

The management of Primary Root Caries is dictated by its differential diagnosis, the basis of which has been outlined in Section II-120. Essentially this has relied upon the clinician's judgement as to whether or not an exposed root surface reveals signs indicating the presence of an active carious process within the root dentine. Presumably this infers the presence of cariogenic micro-organisms within the tissue bringing about its destruction when supplied with acceptable cariogenic substrates. The basis of management would therefore, logically include:

- the attempted elimination of the cariogenic substrate by dietary and oral hygiene measures;

- the elimination of any cariogenic organisms by the topical application of chemo-therapeutic agents from the immediate environment of the root dentine and possibly from any carious dentine;

- attempts to recalcify the partially decalcified dentine matrix through the topical application of suitable agents with hopefully an increased resistance to further decalcification;

- the removal of dentine exhibiting the signs of an active carious process and, containing cariogenic organisms;

- the protection of selected sound root dentine considered to be at particular risk by restorative materials.

In practice, these alternatives result in dentists prescribing one or other of the following options:

- to enhance dietary and oral hygiene preventive measures only;
- to also remove any dentine exhibiting signs of active caries; or,
- in addition, to protect any newly exposed non-carious dentine with restorative material

Chemotherapy is believed to be a treatment strategy that is rarely, if ever, prescribed for the management of Primary Root Caries in dental practice, though some investigatory work has been undertaken.

II-310 Preventive Measures and Plaque Control

Measures aimed at the prevention or the arrest of Primary Root Caries, are mainly based on the elimination of dental plaque from the surfaces of roots and the institution of dietary controls to reduce the frequency and quantity of readily fermentable carbohydrate ingestion (Galan and Lynch, 1994). The role of fluoride, which has been so extensively investigated and its benefits, proved with respect to enamel caries, is by no means clear with regard to Primary Root Caries, though some evidence points to its value. Thus, dentists have in general been

restricted in their ability to institute preventive routines to advising patients on diet and on methods of plaque control.

The mechanical removal of plaque from tooth surfaces, which has been a major platform for the prevention of both dental caries and the periodontal diseases for decades, poses special problems with respect to Primary Root Caries. The surface contours, even on the facial surfaces of teeth and gingivae make toothbrush access to root dentine difficult, not least that lying within a gingival pocket. To remove plaque from the inter-dental surfaces of roots by means of a conventional toothbrush is an impossibility. The customary solution is to advocate the use of dental floss or tape but, whilst the generally convex surfaces of the enamel of the crowns of teeth may well be effectively cleaned by such means, root surfaces pose quite different problems. Virtually all root surfaces, with the possible exception of upper central incisor teeth are concave in the facio-lingual direction, so floss spans such concavities and fails to remove plaque from these surfaces. Only small 'Inter-Space' brushes when used with considerable dexterity are likely to access such surfaces and, therefore, prove to be an effective measure. However, dentists experience considerable difficulty in teaching patients how to effectively use dental floss and to persuade them to do so habitually; and belief that the application of interspace brushes to every inter dental root surface will be achieved is even less plausible.

Because dentine has a Knoop hardness of 68 (Craig and Payton, 1958) in contrast to enamel at 11, the mechanical removal of plaque from its surface inevitably also results in some loss of tissue. Toothbrush abrasion is now a very common phenomenon and invariably relates to the

loss of root dentine from the facial aspects of teeth, especially of canine and premolar teeth of either the upper or lower arch. Consequently, the traditional method of plaque control in the prevention of dental caries creates another problem even when access permits it to be used effectively.

Where Primary Root Caries is already present preventive measures are still instituted (Hellyer and Lynch, 1989b). First, when the clinician judges the lesion to be inactive, in the hope that any further emphasis on prevention is likely to reduce the chance of re-activating the process, and secondly, when a lesion is judged to be actively carious. With the latter, the dentine judged to be heavily infected by cariogenic micro-organisms, invariably on the basis of its Texture, is mechanically removed until the root surface is judged to consist of sound or, non-active carious dentine. It is then treated as though the root dentine had never been carious. Clearly, lesions amenable to such management are relatively superficial though they may be extensive, for then the changed surface morphology is unlikely to be significantly more difficult to maintain plaque free, whilst the placement of a restoration would almost certainly involve the unacceptable destruction of sound tissue and might be remarkably difficult to achieve.

II-320 Restorative Measures

The traditional management of any carious lesion of dentine has involved the excision of infected, necrotic, and partially decalcified dentine and its replacement with biologically acceptable materials with suitable physical and mechanical properties. A very early requirement of student clinicians is that they develop the clinical skills and judgement to expose the

interface between actively carious and inactive or sound dentine by the removal of the first without incurring trauma to the second and, moreover, in the crown without damaging or removing more enamel than is absolutely essential to gain access to the carious dentine beneath. This concern for the conservation of sound dentine is not simply the need to ensure the strength of the caries-free tooth but also, and most importantly, because dentine alone is not a tissue, it is the dentine and the dental pulp together: the pulp-dentine complex, which is affected. Potentially, the cutting of dentine together with the consequences of the carious process can hazard the continuing vitality and integrity of the entire tooth.

With Primary Root Caries, there is invariably a large surface area susceptible to attack and many lesions are extensive, involving more than a single aspect of the surface of a root. The thickness of root-dentine in all teeth is less than the thickness of coronal dentine and on some surfaces, and especially of certain teeth, is very limited indeed. An extreme example would be the inter-dental surfaces of lower incisor teeth. The removal of carious root dentine from such surfaces is clearly a process which puts the dental pulp and, therefore, the whole tooth at great risk; not least since the neuro-vascular bundle supplying coronal pulp-dentine must pass through the narrow root-canal system.

The restoration of teeth from which moderately extensive Primary Root Caries has been removed poses a number of problems. Visibility and isolation from oral fluids (saliva, gingival secretion or haemorrhage) are particular problems, whilst the maintenance of pulpal integrity through the use of biologically acceptable dressings to the pulp-dentine reduce the depth of lesions but do not provide the aesthetic, physical and mechanical

qualities required of a restoration. Furthermore, the placing of any restoration into a cavity in markedly curved surfaces can be technically very difficult. Until the advent of materials such as the glass-ionomer cements, which have a degree of adhesion to dentine, Primary Root Caries lesions needed to be restored with non-adhesive materials including silver amalgam; cast or cohesive gold or fused porcelain with zinc phosphate cement lutes; or silicate cements. All these rely on mechanical means of retention, which can be both traumatic and difficult to achieve in curved surfaces, eg increased depth; undercut or near parallel walls of significant length; or pins. Furthermore, many of these materials exhibit a phenomenon known as 'microleakage' ie gaps are left between the two opposing surfaces of the restoration and the dentine which may predispose to recurrent caries (Kidd, 1976a, b; Taylor and Lynch, 1992). The detrimental effects on the pulp-dentinal complex related to micro-organisms invading these gaps have been documented (Brännström, 1981; Bergenholtz et al, 1982).

Whilst the adhesive materials, when used correctly, result in reduced or complete absence of microleakage, especially with regard to enamel (Lynch et al, 1989a, b; Galan and Lynch, 1993b, c), their adhesive properties are of value mostly when they can be applied to the dentine, which is not the case when dressings such as the calcium hydroxide-based cements need to be used to protect the pulp-dentine. Marginal adaptation (Taylor and Lynch, 1993) can also be a problem. Many authors claim that the material of choice for restoring root caries lesions is the glass-ionomer cement (Wei, 1984; Billings et al, 1985; Kidd, 1989; Titus, 1991). However, the technical problems associated with placing the glass-ionomer cements,

into prepared cavities extending over two or more surfaces of a root are considerable (Tay and Lynch, 1990a, b), and even more so with amalgam. The characteristics of even the glass-ionomer cements have many deficiencies (Wilson and McLean, 1988; Tay and Lynch, 1989a, b; Lynch and Tay 1989a, b, 1991) which makes them far from ideal materials with which to restore root caries lesions. Thus the strategy of managing Primary Root Caries by caries removal and the subsequent restoration of the tooth is far from ideal in terms of efficacy, practicality, prognosis, and cost, not to say that of patient comfort. There is clearly an urgent need to evolve and justify alternative effective strategies for its management.

II-330 Chemotherapeutic Measures

More than fifty years ago attempts were made to convert active dentine caries into inactive or arrested caries by applying solutions to the surfaces of lesions. Usually this related to extensive carious lesions of coronal dentine when the total removal of all carious dentine was likely to result in the exposure of the pulp. Both eugenol and tincture of iodine have been used in the hope that they might be effective, but the agent most frequently employed was a silver nitrate solution which, after its application, would be treated with eugenol to precipitate metallic silver from the silver nitrate wherever it had reached within the decalcified carious dentine. Anecdotal observations of the efficacy of such treatment in preventing further carious activity abound, but it has proved impossible to locate reports of sound research to support the claims that have been made. However, these early attempts at sterilising carious dentine attained only limited acceptance largely due to the fact that the Black staining produced by the precipitated silver was difficult to distinguish from the Black discoloration of many carious lesions themselves and the discoloration of dentine could mimic recurrent caries beneath enamel adjacent to restorations. Clearly the use of silver nitrate on Primary Root Caries was aesthetically unacceptable in almost all circumstances.

II-331 Antibacterial Agents. The increasing appreciation over the years that the two major dental diseases: dental caries and the periodontal diseases, are both dependent upon the activity of micro-organisms, first within dental plaque and then within the tissues themselves, has lead to the development of a number of Chemotherapeutic agents for topical application by means of toothpastes, mouthwashes, gels, or varnishes.

More rarely, the systemic use of drugs has been proposed for the management of some of these diseases. To increase the efficacy of topically-applied agents, devices have more recently been evolved which ensure the slow release of drugs to bring about significantly high levels of an antimicrobial agent at desired locations for extended periods of time.

Agents that have been employed in the management of these oral diseases through their unsupervised routine application, by being incorporated into dentifrices or mouthwashes include (Scheie, 1989; and Marsh 1991 and 1992):

- Bisguanide (eg Chlorhexidine);
- Phenol (eg triclosan);
- Metal Salts such as copper or zinc;
- Essential Oils such as thymol or menthol;
- Plant Extracts (eg Sanguinarine);
- Surfactants such as sodium dodecyl sulphate and
- Quaternary Ammonium Compounds such as cetylpyridinium chloride.

Synergistic interactions between two or more of these drugs have also been claimed to increase the clinical benefits of their use; eg zinc with triclosan. However, whilst there is evidence that such agents reduce plaque levels and gingivitis, there is no clear indication that they beneficially affect Primary Root Caries, though Cummins (1991) states that even at low levels of concentration, they can inhibit bacterial metabolism, including acid production.

The most commonly used agents for topical application to the teeth to prevent or arrest enamel caries are those containing fluoride ions. Bradshaw et al (1990) reported that Mutans streptococci become increasingly sensitive to fluoride ions as the pH falls. It is possible that the routine topical application of fluoride ions will to some extent inhibit the metabolism of such cariogenic organisms.

Goodson (1987) referred to the pharmacokinetic characteristics of drugs used in the prevention or management of oral disease: their toxicity, potency, permeability, intrinsic efficacy, and substantivity, but all these characteristics are affected to varying extents by individual variations in the flow of both saliva and crevicular fluid.

The **toxicity** of a drug needs to be considered with respect to both the consequences of its consumption, since it needs to be used in the mouth, and regarding its toxicity when topically applied to oral tissues. The Merck Index estimates the human fatal doses of the following list of agents (in litres) to be:

- 2.0 percent iodine : 0.05 L
- 0.05 percent Sodium fluoride : 25 L
- 2.0 percent Sodium fluoride : 1.6 L
- 0.005 percent cetylpyridinium : 280 L
- 0.06 percent thymol : 114 L
- 0.12 percent chlorhexidine : 116 L

and even larger amounts of sanguinarine would need to be consumed for a fatal outcome. Clearly iodine and sodium fluoride are the most toxic of the

drugs listed but, at the concentrations used, even sodium fluoride would need to be consumed in considerable quantity before proving fatal. The other four agents, including the frequently used chlorhexidine and thymol, have very low toxicity and it would require huge volumes to be swallowed to produce toxic effects. However, there is the danger that unsupervised home use of fluoride gel over a prolonged period may result in excessive fluoride ingestion, causing gastric irritation (Whitford and Ekstrand, 1988). Tetracycline has been employed topically in the management of some periodontal diseases but its toxicity is considered to be markedly lower than any of the other agents referred to (Merck Index).

Since most of the oral mucosa exhibits non-keratinizing epithelium (except the dorsum of the tongue, the gingivae and the hard palate), the potential toxicity to these tissues of any topically applied agent needs to be considered, for they are capable of easily absorbing them. This applies equally to both the antimicrobial agent itself and the medium in which it is dissolved. However, to date, no adverse effects have been reported following the long-term use of such topically applied drugs (Marsh, 1992).

The **potency** or effectiveness of any antibacterial agent is of great significance. It would seem that, of the topically applied agents reported on, sanguinarine (Dzink and Socransky, 1985), chlorhexidine (Emilson, 1977) and tetracycline (Emilson, 1977) are highly potent, whilst fluoride (Mandell, 1983) and especially cetylpyridinium (Tanzer et al, 1979) and thymol (Evans et al, 1977) have very low potency, as do commonly used inorganic salts such as sodium chloride and sodium bicarbonate. One of the modes of action of chlorhexidine at low concentrations is based on the inhibition of the phosphoenolpyruvate-phosphotransferase sugar transport

system that is used by oral streptococci for acid production (Marsh et al, 1983) and the acidogenicity of dental plaque *in vivo* has been reduced after rinsing with chlorhexidine (Oppermann, 1979; Oppermann and Gjermo, 1980). Though some investigators have questioned the therapeutic value of agents which simply reduce plaque mass, even by as much as 30-50 percent (Kornman, 1986), it would seem logical that the most potent agents are likely to be the more effective in the management of dental caries. Chlorhexidine can also inhibit arginine uptake and catabolism in *Streptococcus sanguis* (Rogers et al, 1987), inhibit proteolysis (Minhas and Greenman, 1989, Millward and Wilson, 1990), affect other membrane functions, including ATP synthase activity and the maintenance of ion gradients in streptococci (Harold et al, 1969), damage cell membranes (Gjermo, 1989), and adhesion of bacteria to tooth surfaces is reduced (Marsh, 1992)

In contrast to systemically administered drugs, a high degree of solubility and membrane permeability may not be desirable characteristics for topically applied antibacterial agents, especially those which have relatively high toxicity, notably sodium fluoride which has a low molecular weight and is readily absorbed, but is high on the toxicity scale. On the other hand chlorhexidine and sanguinarine are both large, highly charged molecules that are poorly absorbed but exhibit low toxicity.

The **intrinsic efficacy** of an antibacterial drug is the fraction (percent) of the maximum achievable effect (ie complete inhibition of growth) that can be obtained. None of the drugs currently in use achieve 100 percent.

When 10 ml of a 0.2 percent solution of chlorhexidine are held in the mouth for one minute, approximately 30 percent is retained (Bonesvoll et al, 1974) but less than 1 percent of a sodium fluoride rinse is retained (Petersson, 1976), chlorhexidine being said to have a high degree of **substantivity** and sodium fluoride a low degree (Van Abbe, 1974). This retention is due to their non-specific binding to sites other than the primary site of drug action as a result of Van der Waals forces, ionic and hydrophobic attraction or covalent bonding. Positively charged chlorhexidine ions can easily adhere to negatively charged proteins such as collagen or salivary mucins (Hjelford et al, 1973; Bonesvoll et al, 1974; Gjermo, 1989). It would seem that agents such as chlorhexidine with a high degree of substantivity would be most effective since they will provide large non-specific reservoirs, but this is clearly not essential since the topical application of fluoride has been demonstrated to reduce enamel caries (Isaac et al, 1958). The strong antimicrobial action of chlorhexidine *in vivo* is most likely to be related to the fact that chlorhexidine is adsorbed by the teeth and mucosal surfaces and is subsequently released in bacteriostatic concentrations over a prolonged period of time (Gjermo et al, 1974; Schaeken, 1984). Other agents, such as cetylpyridinium chloride, which have equivalent antimicrobial activity in the laboratory, produce little antiplaque benefit in humans (Gjermo et al, 1970), possibly because these inhibitors may be inactivated when adsorbed to surfaces (Moran and Addy, 1984).

<u>II-332 Delivery Systems</u> Though supragingival plaque may be successfully controlled by mechanical means (Lindhe and Nyman, 1975) scrupulous oral hygiene is rarely achieved (Svutan et al, 1990). Anti-plaque

agents were therefore developed to supplement mechanical plaque control (Kornman, 1986; van der Ouderaa, 1991). Drugs may be administered by one or other systemic method (ingestion, intravenously or intramuscularly) or topically, which in the mouth means through the use of mouthwashes or toothpastes, or by the application of solutions, gels, sprays or varnishes (Cummins and Creeth, 1992; Gaffar and Afflitto, 1992). Dental plaque and dentine caries do not have blood supplies, as do the periodontium, gingivae, and bone, so it is more appropriate to consider delivering effective antibacterial agents to them via a topical route. However, as referred to above, any topical application system is subject to wide variations in the concentration of a drug at the desired target site, if only due to the wide variations in salivary flow, eg from 60 ml/hr during waking hours to 3 ml/hr or less during sleep, a 20 fold difference. The successful use of use of chlorhexidine solutions and gels for chemical plaque control, applied to the problem of primary coronal caries, has been reviewed by Kidd (1991). However, secondary or recurrent caries is the most common cause of failure of amalgam restorations (Mjör, 1989). Kidd and Joyston-Bechal (1991) showed that chlorhexidine solutions and gels can pass around freshly packed amalgam restorations, and suggested that if it can also be shown that the organisms responsible for the disease are sensitive to chlorhexidine at the concentrations used, then a clinically realistic regimen might be devised for the management of recurrent caries. The suppression of the cariogenic micro-organisms, which are associated with the demineralisation of tooth tissue, results in a reduction in dental caries (Rask et al, 1988). After a short-term intensive treatment of the dentition with chlorhexidine, *S mutans* is suppressed *in vivo* for a significant length of time (Emilson, 1981; Maltz et al, 1981; Zickert et al, 1982; Kristoffersson

and Bradthall, 1982; Schaeken et al, 1984; Schaeken, 1984). Chlorhexidine can reduce dental plaque, caries and gingivitis in humans (Gjermo, 1989; Addy, 1990). It is a broad spectrum antimicrobial agent with activity against a wide range of Gram-positive and Gram-negative supragingival and subgingival plaque bacteria (Emilson, 1977; Stanley et al, 1989) and fungi, including Yeasts. Mutans streptococci are more sensitive to chlorhexidine than other oral streptococci (Emilson, 1977; Stanley et al, 1989) and this has been successfully achieved in plaque and saliva (Schiott et al, 1976; Mikkelsen et al, 1981). *Actinomyces* spp in plaque can also be reduced by chlorhexidine (Briner et al, 1986).

The oral cavity is a convenient container into which solutions may be introduced in an attempt to reach micro-organisms in plaque or within dentine caries, whilst 'washing' all the intra-oral surfaces. Such methods, including rinses, toothpastes, and sprays are 'topical' and do target the oral cavity rather than being 'systemic' and affecting the whole body. However, they are not precisely targeted on plaque and root dentine. More precise targeting has been termed 'controlled' eg the intra-pocket monolithic fibre delivery system of Goodson et al (1983) and the intra-oral fluoride delivery system of Mirth (1980). The benefits in using a 'controlled' delivery system include: an improved pharmacokinetic response; a greater ability to localise the drug adjacent to the disease site; and more control of its local concentration at a lower total dose. Moreover, since the applications of the 'controlled' systems are carried out by professional personnel, patient compliance with a prescribed routine is less variable. The enhanced control over drug concentration at the target site that can be achieved by 'controlled' systems of delivery, compared with more general mouthwash

type systems, has been demonstrated through the use of monolithic intra-pocket tetracycline delivery fibres (Goodson et al, 1983), when gingival fluid concentrations of 400-1000 μg/ml for ten days were achieved. In contrast, Bonesvoll et al (1974); Bonesvoll and Gjermo (1978) and Southard et al (1984) all showed that following the spitting out of mouthwashes, drug concentrations rapidly fell to one-tenth of their initial concentrations and declined exponentially with half-times of 0.5 to 4.0 hours.

II-333 Primary Root Caries 'Controlled' Systems The two main ways in which specific areas of the dentition have been targeted for reasons of the prevention or management of the dental diseases are through the use of either gels or varnishes as media for the active agents. Few attempts have been reported of the development and implementation of such strategies in respect of dentine caries, though more is reported in respect of the prevention and early management of enamel caries.

Gels have been used as media to apply chlorhexidine with or without fluoride ions since 1974. In that year Regolati et al reported that experimental caries in rats was reduced as a consequence of the daily application of gels containing either or both chlorhexidine and sodium fluoride, the greater reduction being obtained by using a chlorhexidine and sodium fluoride gel.

Emilson et al (1976) found that Mutans streptococci counts in human dental plaque were reduced to zero over a three month period when a chlorhexidine / fluoride gel was applied in custom-made trays, and Keltjens et al (1987b) obtained comparable results in root surface plaque covered by overdentures when Mutans streptococci were reduced almost to zero and

the total colony forming unit counts were reduced significantly. In this work gels containing 5 percent chlorhexidine and 0.1 percent NaF were first used daily, later the concentration of chlorhexidine was reduced to 1 percent. Ostela et al (1990) showed that salivary Mutans streptococci counts were reduced by more than 50 percent when a gel containing 1 percent chlorhexidine and 0.2 percent NaF was either applied six times over a two day period or by tooth brushing twice daily for one week. However, the numbers had returned to their original levels after 70 days, presumably due to the recolonisation of accessible surfaces by organisms that had survived in inaccessible fissures or defects. The limited ability of gels to easily access such spaces was demonstrated by Perera (1976).

The initial infection of the mouths of children was investigated by Tenovuo et al (1992). The teeth of mothers were treated with gels containing 1 percent chlorhexidine and 0.2 percent NaF twice a year, commencing when their children were one year old. The appearance of Mutans streptococci in the saliva of the children at two years of age was delayed, but by three and four years there were no delays. However, the caries experiences of the children at four years of age did show a reduction over a control group, dmft's being 0.59 and 0.68, respectively.

In 1990, Keltjens et al reported on the effects on the incidence of root caries of the abutment teeth covered by dentures, as described above, when their surfaces were treated daily with gels containing chlorhexidine (5 percent or 1 percent) and sodium fluoride (1 percent) and found that it had been eliminated by this regime. These observations confirmed the earlier work of Regolati et al (1974) who obtained a reduction in the incidence of experimental caries in rats when gels containing chlorhexidine

and / or sodium fluoride were applied daily to their dentitions, the greatest reductions being when the two substances were combined.

In 1988, Gisselsson et al reported that a 50 percent reduction in the incidence of approximal carious lesions had been achieved when 1 percent chlorhexidine gels were applied to the dentitions of children four times a year, these were applied from syringes and then spread by flossing.

Varnishes Caries prevention through the application of fluoride containing varnishes to the enamel of teeth (De Bruyn and Arends, 1987) which can adhere to teeth for periods of several weeks and the promise of a beneficial effect of the topical application of anti-cariogenic agents or microbiological counts on caries incidence led Balanyk and Sandham (1985) to investigate the incorporation of other agents into a varnish, and other researchers have followed similar pathways. Sandham and colleagues (1988) used a varnish incorporating 20 percent by wt chlorhexidine which was applied to the teeth on average 2.7 times each week over a mean period of 34 weeks and claimed the total eradication of salivary Mutans streptococci. Only 21 subjects were involved in this study and in twelve of them, in which about five applications per week were made, the results were recorded as being unsuccessful. However, further data reported three years later (Sandham et al, 1991) did show some promise. Sandham et al used a silane fluoride varnish (Fluor Protector)[1] as the vehicle for the chlorhexidine, which Seppa et al (1982) had shown to be only weakly caries preventive, so it is probable that any beneficial result obtained was indeed due to the chlorhexidine.

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[1]Vivacare, Vivadent, Liechtenstein

Schaeken and De Haan (1989) investigated the effects of varnishes containing remarkably high concentrations of chlorhexidine (50 wt percent) and smaller quantities of sodium fluoride (2.5 percent or 5.0 percent) either alone or in combination. They reported about a 50 percent reduction in the incidence of Primary Root Caries when varnishes containing either of these agents were applied to susceptible surfaces every third month and a 33 percent reduction in the Mutans streptococci count from plaque when the chlorhexidine varnish was used. In later studies (Schaeken et al, 1991a), the chlorhexidine in the varnish was reduced from 50 percent by wt to 40 percent by wt to reduce undesirable effects on the soft tissues. Two different strategies were compared for their effects on suppressing the Mutans streptococci counts from plaque: for one group the varnish was left *in situ* for a period of two to three months after being applied to approximal tooth surfaces; in the other group it was removed 15 minutes after application. No differences were detected between the two groups.

Hilderbrandt et al (1992) have used a different approach to the use of chlorhexidine varnish. Individually fabricated mouthguards were coated internally with a varnish containing 3 percent chlorhexidine and worn by patients for seven hours each night for seven consecutive nights. Approximately 50 percent reduction in salivary Mutans streptococci was achieved immediately after this regime and a slight reduction was still detectable three months later, but this latter effect was associated with a significant rise in the salivary *Actinomyces ssp* counts, the significance of which is not yet known.

Fure and Emilson (1990) studied the suppression of Mutans streptococci on root surfaces using chlorhexidine gel supplemented with

chlorhexidine varnish treatments and demonstrated an additional effect of the varnish application.

It would seem that the use of varnishes containing anti-cariogenic agents as media, targeting defined sites, shows some promise, not least since only a few applications might be required annually. However, investigations are still required to identify optimum regimes, and to date no studies have been reported on the effects of such varnishes on the microbiology of carious dentine itself. Goho and Aaron (1992) have claimed that the application of a chlorhexidine varnish to the cut dentine in cavities prepared in extracted teeth increased the antimicrobial properties of the final restorations when a varnish was applied. Antimicrobial agents can behave very differently *in vivo* compared to *in vitro* due to many factors including; the exposure time required by the antimicrobial agent, whether this agent has microbicidal or microbistatic effects, the penetration of these inhibitors into lesions or plaque, and the environmental conditions within lesions or plaque. Consequently, *in vivo* testing of antimicrobial agents is required to test efficacy.

Antibacterial agents have been combined in varnishes as they have been in other media, notably chlorhexidine with thymol, and Huizinga (1991) observed an enhanced effect *in vitro* from a combination of these when compared with either of them individually, in a varnish called 'Cervitec'. Schaeken and De Haan (1989) after *in vitro* studies, reported that *S sanguis* was inhibited by a varnish containing both chlorhexidine and thymol but not when thymol was excluded. Lactobacilli strains are relatively resistant to chlorhexidine (Zickert et al, 1982; Cleghorn and Bowden, 1989) compared with *S mutans* (Maltz et al, 1981). Cervitec showed antimicrobial

activity against Lactobacilli, Streptococci, Actinomyces and Yeasts *in vitro* (Petersson et al, 1991) which ties in with data obtained. *in vivo* showing that these antimicrobial agents, chlorhexidine and thymol can leak out from this polymer based varnish (Huizinga et al, 1991; Petersson et al, 1991; Marsh, 1992) Thymol, along with menthol and eucalyptol is known as an 'Essential Oil', an extract of certain plants and chemically it is the hydrocarbon monohydroxyphenol: $C_{10}H_{14}O$ (Kato et al, 1990; Marsh, 1991) and as long ago as 1890, Miller discussed the antiseptic effect of thymol. These oils have been used in traditional Chinese medicine for centuries and one extract of the plant Mosia Cheninensis which contains 22 percent thymol has been shown to possess antimicrobial properties against Mutans streptococci from plaque (Osawa et al, 1990). Thymol has a high antimicrobial activity compared to the other essential oils (Hedgecook, 1967). The commercial mouthwash 'Listerine'[2] is essentially a tincture of essential oils, including thymol, which is believed to be a most effective agent against a range of plaque micro-organisms (Gordan et al, 1985; Axelsson and Lindhe, 1987) including candida albicans (Kato et al, 1990), though no report of the efficacy of the different individual agents has been made. Walker (1988) reported on studies of the effect of 'Listerine' on periodontal disease and claimed 39-48 percent reductions in existing plaque levels and 45-56 percent reductions in the development of new plaque. 'Listerine' diluted four times has been shown to be effective against *S mutans* and *S sobrinus* and at eight times dilution against Lactobacilli casei, ie 10-50 percent reduction in numbers following exposures of 10-30 seconds (Kato et al, 1990). Lamster et al (1983) believe that this is brought about by the bacterial protein denaturation in cell walls resulting in the

[2]Warner-Lambert Co, Morris Plains, NJ, USA

leakage of intra-cellular elements. No significant adverse reactions to the use of 'Listerine' have been reported though some individuals object to its taste. Thymol would seem to be a weak sensitizer but needs to be in contact with inflamed skin to produce signs of dermatitis and even such a sensitised patient was able to use thymol orally without experiencing any reaction (Fisher, 1989). Thymol and chlorhexidine have been accepted by the American Dental Association's Council of Dental Therapeutics (1985) for the control of supragingival dental plaque and gingivitis and are the only two agents accepted by this Council (Mandel, 1988).

Varnishes appear to release thymol and chlorhexidine simultaneously, this release is initially rapid but then it slows and continues for up to three months with chlorhexidine being the more persistently released (Huizinga, 1991). One such varnish, 'Cervitec'[3], has been shown to release 1 $\mu g/cm^2$/day from the surfaces of root dentine to which it had been applied *in vitro*, the dentine acting as a depot enabling the drug to be released over a six month period (Arends and Ruben, 1993). This supported the observations made by Petersson et al (1991) who found salivary Mutans streptococci counts were still significantly reduced three months after an application of this varnish to root surfaces in a clinical trial. Huizinga et al (1990) have reported that Cervitec reduces root demineralisation *in situ*. Such observations have lead Ten Cate et al (1993) to state that its use in the prevention of Primary Root Caries is indicated.

The media, or solvents containing 1 percent by wt chlorhexidine and 1 percent by wt thymol in 'Cervitec' varnish are ethanol and ethylacetate. *In*

[3]Vivacare, Vivadent, Liechtenstein

vitro experiments designed to determine whether these solvents possessed any significant anti-microbial properties when used in this way surprisingly failed to show any. 'Cervitec' varnish was compared with a placebo 'Cervitec' varnish, which contained neither phenol nor chlorhexidine, and the latter produced no desirable antimicrobial consequences (Petersson et al, 1991).

Varnishes containing antimicrobial agents should permit a localised targetted effect to the selected sites *in vivo* whilst preserving microbial homeostasis in the mouth. One of the unfortunate side-effects of the intra-oral use of chlorhexidine is the staining of teeth that can occur, another is an altered taste perception (Fløtra et al, 1971), but the low amount used locally of this drug in 'Cervitec' varnish almost certainly ensures that these side effects are unlikely to occur and, to date, they have not been reported. The odour of the varnish has been commented upon, though it has not been perceived as being offensive. It would therefore, seem that a regime involving the use of 'Cervitec' varnish would almost certainly be perceived as both convenient and acceptable to patients.

II-400 Conclusions

This work is concerned with the factors which might contribute to the reliable differential diagnosis of Primary Root Caries, for only with such a foundation can clinicians and epidemiologists have confidence in their conclusions. Beyond diagnosis lies the potential for the prevention of the disease and the most appropriate clinical intervention, whether by medicaments or various restorative procedures. The morphology of the human dentition is such that root surfaces may be easily accessible to direct vision and tactile investigation eg the labial aspect of upper canine teeth exhibiting some gingival recession but no periodontal pocketing. However, other surfaces within deep periodontal pockets; the approximal surfaces of teeth in a crowded dentition; the lingual aspects of lower molar teeth protected by a powerful tongue; or the buccal aspects of upper molar teeth made inaccessible by the ascending ramus of the mandible; are a different matter. Vision alone cannot be relied upon in the great majority of situations.

The fact that the structure of dentine is so much in contrast with that of enamel, means that exposed surfaces can be of many very different hues due to the absorption of extrinsic stains on to its structure or the exposure through abrasion of ever deeper layers of dentine with their different appearances. Vision alone cannot be considered to be a reliable diagnostic criterion, it can only be an indication of the need for further investigation.

The differential diagnosis of incipient, arrested, and active Primary Root Caries and abrasion or erosion would seem to depend more than

anything else on the microbiological characteristics they display. Colour change, Contour, Location on the root surface, age or periodontal condition either singly or collectively would seem to be inadequate criteria for reliable diagnoses. By far the most promising of the clinical signs of Primary Root Caries available would seem to be the changes in the Texture of root dentine that occur. Therefore, well founded research would seem to be a top priority to relate the microbiological characteristics of lesions and their clinical signs and symptoms, especially Texture, if reliable criteria for the assessment and management of Primary Root Caries are to evolve.

There is ample evidence to indicate that, in spite of the development of new restorative materials and a heightened awareness of the ever increasing problems of Primary Root Caries, existing management strategies are unlikely to come near to proving adequate. Strategies which do not rely so heavily on unrealistic assumptions of patients' abilities to control plaque and dietary factors, as do the present ones, and on interventions by dentists which do not rely so heavily on mechanical techniques, urgently need to be proven. Such changes in the philosophical and technical approaches to the management of Primary Root Caries will need to be based on sound research which ties together the clinical and the microbiological characteristics of the disease itself and the effects of preventive and non-destructive treatment strategies on them. For example, if the pathogenic capacity of the cariogenic micro-organisms in Primary Root Caries could be decreased, and if the bacteriostatic action of low chlorhexidine concentrations can be shown to be long lasting, then the glycolytic function and acid production of the cariogenic microflora should

be reduced thereby permitting remineralisation to occur and a stable condition to be attained.

III HYPOTHESIS

That the Texture of root dentine affected by the Primary Root Caries process is directly related to its level of activity as indicated by its microbiological characteristics; that other signs or symptoms, notably Colour changes or modifications to the surface contour of roots, whilst indicating the wisdom of assessing its Texture, are in themselves unreliable indicators of activity levels but collectively certain clinical signs can prove to be reliable indicators; and that selected topically applied Chemotherapeutic agents might well prove to be convenient, effective, cheap and conservative aids in its prevention and management.

IV MATERIALS AND METHODS

IV-100 Introduction

To test the stated hypothesis, it was decided that Primary Root Caries would be identified *in vivo* and the clinical characteristics associated with each lesion recorded using a standard format. Bacteriological samples would be obtained in each case; cultured using the most appropriate methods available; and the numbers, types and proportions of organisms present would be identified. The clinical and microbiological characteristics of each lesion would then be related to each other. Using this basic methodology, the effects on the micro-flora of Primary Root Caries, of employing a topically applied Chemotherapeutic agent would be determined.

Prior to the commencement of the Chemotherapy study, ethical approval was obtained from the local Ethics Committee of The London Hospital Medical College and Tower Hamlets Area Health Authority (Reference 92/110). Written informed consent was obtained from each individual included in this study after the introductory letter had been presented to each patient.

IV-200 Subjects

The author exclusively recruited suitable subjects to participate in the investigation. All participants were patients of The Royal London Hospital, London E1 2LR who were attending its Dental Institute for routine oral health care. Each subject had given their informed consent for both dental examinations to be undertaken and for samples of Primary Root Caries to be taken for bacteriological investigation. Patients with xerostomia based on obvious clinical grounds (Kitamura et al, 1986); those who had undertaken courses of systemic antibiotic therapy or had used anti-bacterial mouthwashes during the previous four weeks; and any who had undergone periodontal surgery within the previous six months were excluded from the study. No lesions covered by partial dentures were sampled (Wright et al, 1992).

The data with which this whole work is concerned has been obtained from a total of 610 Primary Root Caries lesions in 303 patients. The results of the major study undertaken recorded in Sections V-200 to V-600 inclusive, relate to data collected from 169 adult patients (M: 93, F: 76) with an age range of 29 to 80 years (mean ± standard deviation = 56.4 ± 14) and involving a total of 447 lesions (up to a maximum of 11 per patient).

For the study, which compares the microbiological characteristics of Primary Root Caries and the dental plaque overlying them, for which the results are given in Section V-700, 52 patients were recruited (33: M, 19: F) with an age range of 40 to 78 years (mean ± standard deviation = 53.7 ± 12.2); 81 lesions were examined of which 64 were designated clinically as Soft and 17 as Leathery and deemed to require a restoration (up to a maximum of 3 per patient).

The results of the investigations on the effects of Chemotherapy given in Section V-800 were obtained from a further 82 patients (M: 45, F: 37) with an age range of 39 to 75 years (mean ± standard deviation = 54.6 ± 13.2) and involving 82 lesions, of which 40 were designated clinically as Soft and 42 as Leathery but all were identified as needing restoration on the basis of the clinical criteria described under V-310; 20 Soft lesions and 22 Leathery lesions were randomly identified as a study group to be subjected to Chemotherapy, the other 20 Soft and 20 Leathery lesions as a group not to receive any Chemotherapy.

IV-300 Methods and Materials

IV-310 Clinical Methods

The clinical phase of the investigation had three component parts: the recording of the clinical signs of each lesion; the defining of the Treatment Needs which these signs indicated; and the bacteriological sampling relevant to each lesion. All these stages for all the subjects included in the study were carried out by the author.

IV-311 Clinical Signs were of three types:

- Colour. Carefully prepared colour photographs of Primary Root Caries were obtained and from these a four shade guide created. These shades were designated 'Yellow'; 'Light Brown'; 'Dark Brown'; and 'Black' and they were used as the standard when determining the Colour of each lesion being investigated.

- Dimensions. A standard periodontal probe marked at 1 mm intervals was used to determine the dimensions of each lesion. The maximum occluso-gingival (height) and mesio-distal or labio/bucco-lingual/palatal (width) dimensions were assessed as well as the minimum distance from the gingival margin of the lesion and the crest of the gingivae itself. The product of the first two figures was used as an indicator of its overall Size. In addition an estimate of the amount of dentine that had been lost (Cavitation or greatest loss of surface contour) was made by recording the greatest distance between the existing surface of the lesion and what was judged to have been the original root surface.

- Texture. Three categories for the Texture of each carious lesion were defined:

  - 'Hard' required the caries to be comparable to the surrounding sound root dentine;

  - 'Leathery' caries permitted a new No 6 probe[4] to penetrate the surface under moderate pressure and there was some resistance to its withdrawal;

  - 'Soft' lesions permitted a new No 6 probe to penetrate the surface with ease and there was no resistance to its withdrawal.

The reproducibility of the probing force used with the new No 6 probe was assessed by mounting 50 teeth (one at a time) with Primary Root Caries lesions onto a Mettler PC 180 micro-balance[5]. Mounting was achieved by using an addition-cured silicone impression putty[6]. The Primary Root Caries lesions were then probed with a 'modest' pressure 50 times and a force of 102.1 ± 6.72 gm was measured (mean ± SD).

Unweighted Kappa (Cohen, 1960) and weighted Kappa (Cicchetti, 1976) have been used to consider areas of total agreement and also to take account of 'near-misses' respectively. At least 40 teeth have been examined on several occasions for Colour, Texture and Perceived Treatment Need of Primary Root Caries lesions and repeated at least one week later. Scores for unweighted and weighted Kappa have exceeded 0.8

[4]Claudius Ash Sons & Co Limited, Potters Bar, Herts, UK
[5]Mettler Instrumente AG, Zurich, Switzerland
[6]Extrude Putty, Kerr, USA

on each occasion indicating at least good agreement for Colour, Texture and Perceived Treatment Need.

The repeatability of measuring the dimensions (Width, Height and Cavitation) of Primary Root Caries lesions have been assessed by examining at least 40 teeth on several occasions and repeated one week later. At least 95 percent of these scores differed by 1.0 mm or less on each occasion indicating reasonable level of agreement (Bland and Altman, 1986).

IV-312 Perceived Treatment Needs were defined strictly in relation to the clinical signs as defined above, these being:

- None: All 'Hard' lesions.

- Chemotherapeutically: 'Leathery' lesions which were considered to be small, easily cleansable and approaching a 'Hard' Texture.

- Carious dentine to be removed: 'Leathery' lesions which were judged to be shallow and the surface of the exposed sound dentine easily maintained plaque free.

- Carious dentine to be removed and the tooth restored: All 'Soft' lesions; 'Leathery' lesions judged to be in surfaces which were difficult to maintain plaque free; and large, cavitated 'Leathery' lesions where pulpal integrity was judged to be at risk.

IV-313 Chemotherapy. Each of the lesions in the two test groups defined in Section V-200 for Management by Chemotherapy (results in Section V-800) were first cleansed of supra-gingival plaque using a sterile toothbrush and sterile distilled water; the teeth were then isolated using cotton-wool rolls; dried with air using a dental 3:1 syringe for 10 seconds, then varnished.

'Cervitec Varnish'[3] was employed, constituted of:

- Chlorhexidine	1 percent W/W
- Thymol	1 percent W/W
- Ethyl acetate	48 percent W/W
- Ethanol	40 percent W/W
- Polyvinylbutyrol	10 percent W/W

After application and evaporation of the solvents, the Varnish contains the equivalent of 6.5 percent of both chlorhexidine and of thymol (Arends and Ruben, 1993). Each Primary Root Caries lesion and the surrounding sound tooth surfaces were varnished using a disposable brush; the varnish solvent rapidly evaporated permitting a total of five coats to be applied in rapid succession. Patients were then instructed to avoid normal tooth-brushing for a minimum of four hours, but otherwise to follow their usual custom, so as to retain the thin layer of varnish undisturbed. Biopsies were obtained (see IV-314 below) of each lesion 24 hours after the application of the varnish.

IV-314 Biopsies. Two categories of material were obtained for bacteriological examination. For the 81 Primary Root Caries lesions included in the study comparing plaque and dentine (Section V-700) samples of supra-gingival plaque were obtained from the surface of the Primary Root Caries. A sterile, 'flat-plastic' dental instrument was used to lift this from the tooth surface by traversing the lesion in line with the long axis of the tooth across the maximum gingivo-occlusal dimension and the sample was immediately placed in 1 ml of 'Fastidious Anaerobe Broth' (FAB)[7] . The blunt flat-plastic instrument was selected and used with care to ensure that any underlying altered dentine was not removed to contaminate the plaque sample retrieved.

Biopsies of carious dentine were obtained from all the 610 Primary Root Caries lesions studied. Root surfaces were first cleansed to remove plaque and any other material which might contaminate the sample of carious dentine. A hand-held standard fine nylon fibre sterile toothbrush was used with sterile water as a lubricant to cleanse the surface but to avoid the removal of any surface carious dentine. The tooth was then isolated using cotton wool rolls and any moisture removed from the surface of the lesion with a dry sterile cotton wool roll. A new, sterile, Ash 220/221 dental excavator[8] was used to remove a sample of the carious dentine. Pressure comparable to that used in normal clinical practice was used and, so far as was possible, this was standardised on each sampling. The excavator blade was used to traverse the lesion in line with the long axis of the tooth across the maximum gingivo-occlusal dimension. Each biopsy

[7]Lab M, Bury, Lancs, UK
[8]Claudius Ash Sons & Co Limited, Potters Bar, Herts, UK

was immediately placed into 1 ml of Fastidious Anaerobe Broth (FAB)[9] and forwarded to the laboratory within 30 minutes for bacteriological investigation.

The reproducibility of the force used by the 'Flat-plastic' when sampling the supra-gingival plaque was assessed by mounting 50 freshly extracted teeth with Primary Root Caries lesions onto a Mettler PC 180 micro-balance[10]. Mounting was achieved by using on addition-cured silicone impression putty[11]. On each occasion that samples of plaque were taken, forces less than 5 gms were recorded .

The reproducibility of the force used with a new Ash 220/221 dental excavator[12] was assessed by applying pressure, comparable to that used in normal clinical practice, to biopsy the Primary Root Caries lesion in each of 30 teeth. Each tooth was again mounted in an addition-cured silicone putty[11] on the Mettler PC 180 micro-balance[10]. A force of 160.3 ± 16.21 gm was measured (mean ± standard deviation).

IV-320 Laboratory Methods

IV-321 Sample Processing. To each 1 ml of FAB containing a biopsy of carious dentine sterile 3.5 to 4.5 mm diameter glass beads[13] were added. They were then vortexed for 15 seconds to facilitate the extraction of any micro-organisms from the carious dentine and disperse any

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[9]Lab M, Bury, Lancs, UK
[10]Mettler Instrumente AG, Zurich, Switzerland
[11]Extrude Putty, Kerr, USA
[12]Claudius Ash Sons & Co Limited, Potters Bar, Herts, UK
[13]BDH Limited, Poole, Dorset, UK

aggregates, then decimally diluted with FAB, 100 ml aliquots of these being spread as appropriate onto a range of culture media in plates.

- Mutans streptococci (*S mutans* and *S sobrinus*) were enumerated on Mitis-Salivarius Agar[14] supplemented with 0.2 units per ml bacitracin and 15 percent (W/V) sucrose, MSB (Gold et al, 1973) in an anaerobic chamber[15] at 37°C for three days, the total number of these organisms in each sample being determined by counting the colonies identified on each plate. Their identities were confirmed by subjecting up to five of these colonies to further examination by using a set of fermentation and enzymic tests. Tests for the production of acid from N-acetylglucosamine, arbutin and melibose, as well as the presence of α-alactosidase and α-glucosidase activities were useful in differentiating these species (Beighton et al, 1991b).

- Lactobacilli were grown on Lactobacillus Selective Agar LBS[16] in an anaerobic chamber[15] at 37°C for three days, the total number of organisms being determined by counting the typical colonies grown. They were confirmed as Gram-positive, catalase-negative, and unbranched rods (Beighton et al, 1991b).

- Yeasts were grown on Sabouraud Dextrose Agar[17] incubated in air at 37°C for two days, the number of organisms being determined by counting the number of typical colonies on each plate. They

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[14]Difco Laboratories, Teddington, Surrey, UK
[15]Don Whitley, Shipley, West Yorkshire, UK
[16]Oxoid Limited, Basingstoke, Hampshire, UK
[17]Oxoid Limited, Basingstoke, Hampshire, UK

were confirmed as large, ovoid Gram-positive catalase-positive cells (Beighton et al, 1991c).

- Gram-positive pleomorphic rods (GPPR's) were grown on Fastidious Anaerobic Agar[7] supplemented with 5 percent (V/V) horse blood (FAA, LabM) in an anaerobic chamber[15] at 37°C for seven days, the number of organisms, including *Actinomyces spp* being determined by examination of Gram-stained smears of colonies on the Fastidious Anaerobic Agar plates. The number of each colony type was counted and representatives of each examined so that the numbers of Gram-positive pleomorphic rods in each sample could be calculated. The majority of these isolates were *Actinomyces spp*, since they produced acetic, lactic and succinic acids from the fermentation of glucose in peptone - yeast extract - glucose broth supplemented with 0.01 percent Tween 80. Because of the difficulties associated with the speciation of the Gram-positive pleomorphic rods, especially the identification of *Actinomyces spp* (Johnson et al, 1990) all Gram-positive pleomorphic rods were grouped together as a single taxon. From these plates the total numbers of all colony forming units were also determined.

The detection limit for Mutans streptococci, Yeasts and Lactobacilli was 10 cfu per sample, and for the Gram-positive pleomorphic rods, it was approximately 0.2 percent of the number of Colony forming units present in a sample. If no colonies of a given taxon were recovered from a sample, a value of nought was included in the analysis.

IV-322 Data Analysis. The total numbers of colony forming units per sample biopsy were determined by translating the numbers of colony forming units grown on the FAA plates through the dilutions that had been employed to relate them to the total sample. These were transformed to Log_{10} (colony count + 1) to normalise the distributions of the individual colony counts. The numbers of each of the various categories of organisms per sample were determined in the same way and the proportions of all organisms in each sample also expressed as a percentage of the total colony count on the FAA plates. Means and standard errors of values were also calculated, mean values being compared by the Student's paired t-test and by one way analysis of variance using Duncan's multiple range test and Pearson's correlation coefficients were calculated. Distributions were analysed by the Chi^2 statistic(s). The Frequency of isolation of individual taxa from each Primary Root Caries lesion was recorded as zero if fewer than 10 Colony forming units of that particular taxon were recovered from the biopsy. All statistical analyses were performed with the comprehensive statistical suite of programmes: SPSS/PC + V 3.0[18].

IV-330 Reproducibility of the microbiological enumeration from the lesion samples

The degree of reproducibility in the enumeration of micro-organisms from root caries dentine samples was investigated by enumerating the number of individual taxa in 30 individual lesion samples. These root lesion samples consisted of 10 Soft, 10 Leathery and 10 Hard. Each was processed for the total number of bacteria, the number of Gram positive pleomorphic rods, Mutans streptococci, Lactobacilli and Yeasts as described previously.

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[18]SPSS Inc, Chicago, Illinois, USA

Each sample was diluted in FAB twice and each of the two sets of dilutions for each sample were spread onto the appropriate selective and non-selective media. The plates were incubated as appropriate and the number of each taxa was counted from these plates. From these counts the number of the individual microbial taxa in each sample was determined twice.

The reproducibility of these counts was investigated by comparing the 30 pairs of counts for each taxa using the Student's paired t-test and by calculating Pearson's correlation coefficient. These data are summarised as follows:

Micro-organism	Sample 1	Sample 2	r
Total CFU	5.61 ± 1.9	5.70 ± 1.8	0.87 ($P < 0.001$)
GPPR	5.10 ± 1.7	5.25 ± 1.8	0.80 ($P < 0.001$)
Mutans streptococci	3.10 ± 2.8	3.50 ± 2.7	0.64 ($P < 0.001$)
Lactobacilli	2.98 ± 1.8	2.68 ± 2.1	0.75 ($P < 0.001$)
Yeasts	2.20 ± 3.4	2.50 ± 3.6	0.84 ($P < 0.001$)

These results clearly indicate that the microbiological counting methods were reproducible as none of the pairs of data were significantly different ($P > 0.5$) and each set was significantly correlated ($P < 0.001$).

IV-340 Discussion of Methods

Microbiological investigations of root caries in humans can be divided into those designed to determine relationships between salivary levels of caries-associated micro-organisms (particularly Mutans streptococci, Lactobacilli and Yeasts) and root caries prevalence or incidence (Ravald and Hamp, 1981; Billings et al, 1985; Ravald et al, 1986; Fure et al, 1987; Emilson et al, 1988; van Houte et al, 1990; Beighton et al, 1991c; Scheinin et al, 1992); those designed to determine the relationship between the bacterial composition of root-surface plaque on sound root surfaces and the future caries status of those surfaces (Ellen et al,

1985a, b; Emilson et al, 1988); and other studies investigating the relationship between the caries status of root surfaces and composition of bacterial samples that usually represent a mixture of supragingival plaque overlying the lesion and infected dentine intimately associated with the root caries lesions (Syed et al, 1975; Brown et al, 1986; Bowden et al, 1990). The present method differs significantly from these earlier studies since the supragingival plaque was removed prior to taking the dentine sample from an individual lesion. This routine was followed, not least in view of the work of Bowden et al (1990) who removed plaque from "the entire surface of an intact root or lesion" with a scaler recovering approximately 10^9 viable cells, irrespective of the caries status of the root. In contrast, in a pilot study to the present work, the number of Colony forming units recovered from the clinically most severe Primary Root Caries was approximately 10^7, while $<10^3$ Colony forming units were recovered from the surface of Hard lesions. The difference appeared to be due to root surface plaque, sampled by Bowden et al, which was not necessarily associated with lesions.

Crucial to the method of sampling in the present study are believed to have been the standardised procedures used to prepare root surfaces beforehand and the method employed to obtain the samples. In this study comparisons could therefore be made between the total number of bacteria recovered from individual lesions and these could be related to the types of Primary Root Caries lesions classified using a range of independent clinical indices. In other studies the percentage composition of samples was usually calculated and these have either shown no difference between the proportions of Mutans streptococci or Lactobacilli from sound and carious root surfaces (Keltjens et al, 1987a; Emilson et al, 1988; Bowden et al, 1990) or reported significantly greater proportions of these bacteria from carious lesions (Brown et al, 1986; Keltjens et al, 1987a). It is felt that the method employed in this study helps clarify these issues.

V RESULTS

V-100 Introduction

The results of these investigations are presented in seven main sections V-200 to V-800 and, for reasons of clarity, as histograms, in each of which only two variables are related, eg The Colours of Lesions v Their Perceived Treatment Needs. Brief comments are made on each histogram as appropriate. Even so, there are more than 40 such histograms and to further increase this number by presenting the data relating to three or more variables, eg the Colours, Textures, and Microbiology together is believed to be unhelpful for, on such a programme some hundreds of histograms would be involved. The sections are:

- The Inter-relationships of the Clinical Signs of Lesions
  - 11 histograms in V-200

- The Clinical Signs of Lesions and their Perceived Treatment Needs
  - 5 histograms in V-300

- The Clinical Signs of Lesions Related to the Microbiology of Carious Dentine
  - 15 histograms in V-400

- The Perceived Treatment Needs Related to the Microbiology of Carious Dentine
  - 3 histograms in V-500

- Correlation, Discriminant and Regression Analysis of the Clinical Signs, the Perceived Treatment Needs and the Microbiology of Carious Dentine
  - 2 histograms and 5 tables in V-600

- The Microbiology of Carious Dentine Related to the Microbiology of the Overlying Plaque
  - 3 histograms in V-700

- The Microbiology of Carious Dentine Subjected to Chemotherapy Related to that of Controls
  - 6 histograms in V-800

V-200 The Inter-relationships of the Clinical Signs of Lesions

These data are presented in five sub-groups, the 11 histograms displaying the relationships of each variable with the others.

- The Colours of Lesions - 4 histograms in V-210
- The Textures of Lesions - 3 histograms in V-220
- The Locations of Lesions - 2 histograms in V-230
- The Cavitations of Lesions - 1 histogram in V-240
- The Heights and Widths of Lesions - 1 histogram in V-230

In view of the direct relationships of the Sizes and the Widths and Heights of lesions there is no need to relate these, other than in the last of these histograms.

V-210 The Colours of Lesions

The relationships of the Colours of Lesions to the other four clinical signs are displayed in this section, each on a separate page.

Four groups of data are presented in this section, ie relating to:

- the Textures of Lesions: V-211,
- their Locations: V-212,
- their Cavitations: V-213 and
- their Sizes: V-214.

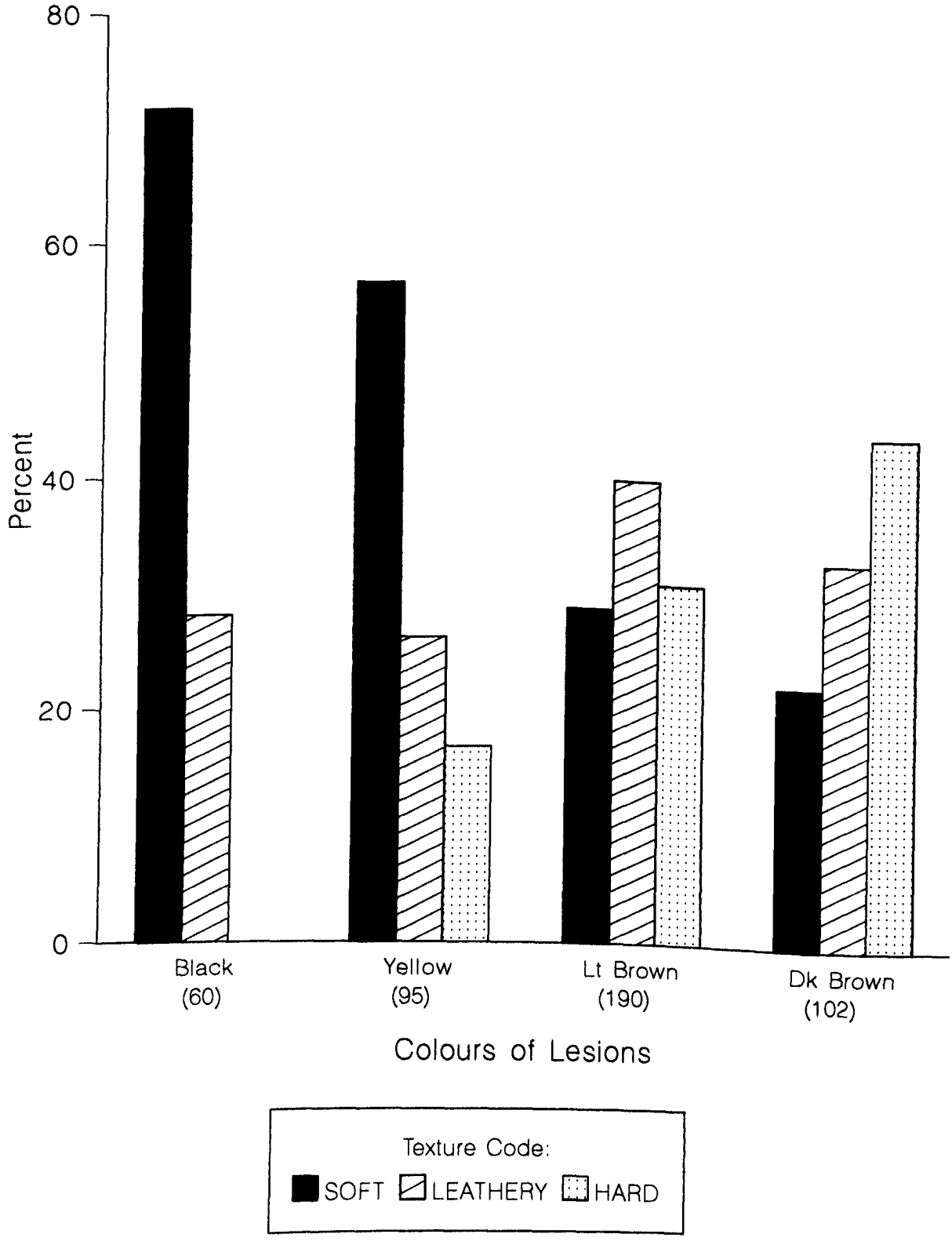
The four categories of Colours are:

- Black; Yellow; Light Brown; and Dark Brown.

The number of Lesions in each Colour group were diagnosed as follows:

- 60 Lesions were Black,
- 95 Lesions as Yellow,
- 190 Lesions as Light Brown and
- 102 Lesions as Dark Brown

V-211
The Colours of Lesions
V
Their Textures

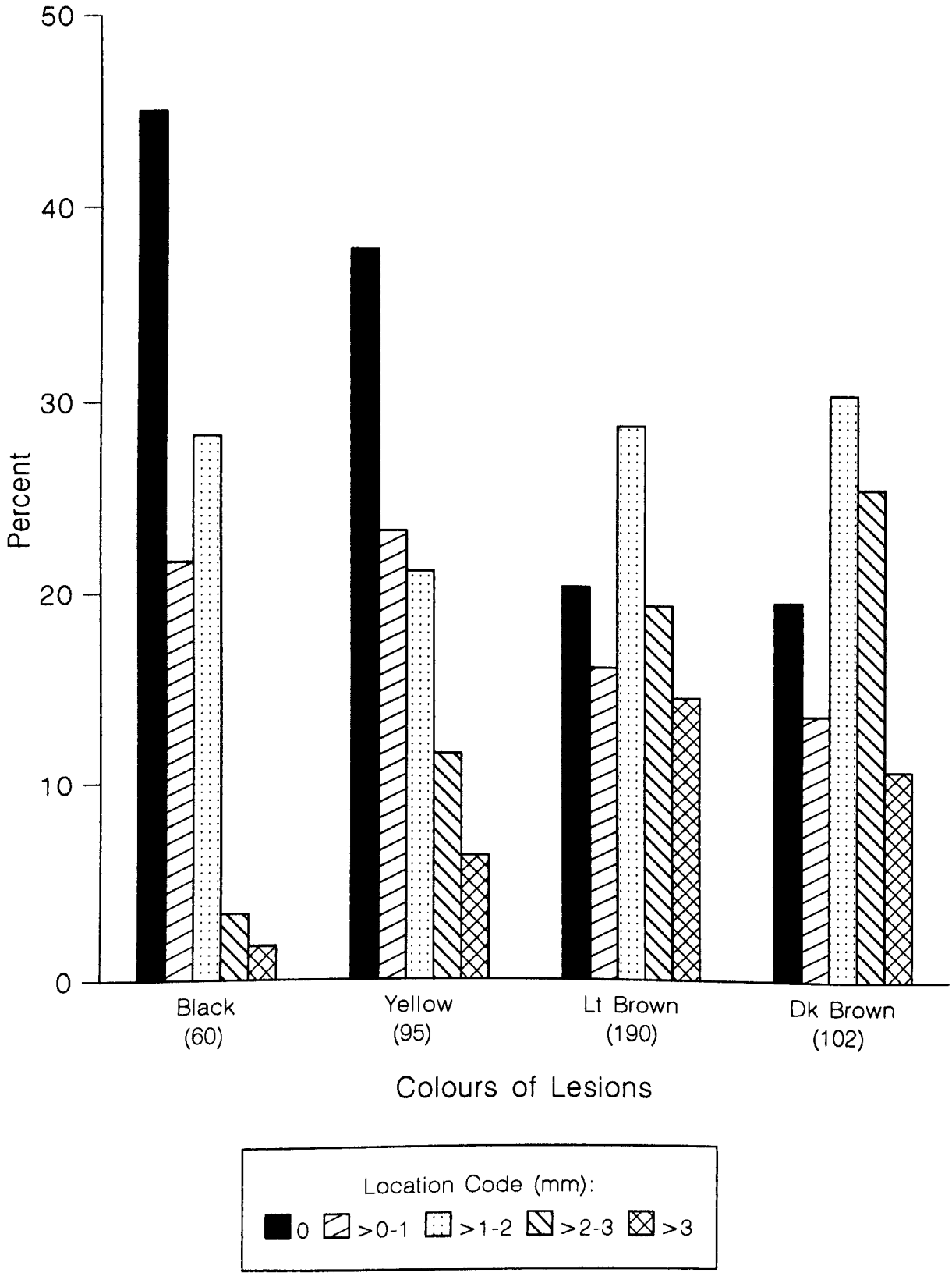


NB. The numbers in brackets indicate the number of lesions

V-211 The Colours of Lesions v Their Textures

The four Colours of Primary Root Caries (Black, Yellow, Light Brown and Dark Brown) are shown related to the three Textures defined as Soft; Leathery; or Hard. Some 71.7 percent of all Black lesions were designated as Soft and 56.8 percent of Yellow lesions were also Soft, but relatively small proportions of Light Brown (28.9 percent) or Dark Brown (22.5 percent) were judged to be Soft. No Black lesions were diagnosed as being Hard whilst the proportions of Hard lesions increased from Yellow (16.8 percent) to Light Brown (31.1 percent) to Dark Brown (44.1 percent). Leathery lesions may, apparently, be any Colour for the proportions of those designated to each of the four Colour categories vary only between Black (28.3 percent); Yellow (26.3 percent); Light Brown (40 percent) and Dark Brown (33.3 percent).

V-212
The Colours of Lesions
V
Their Locations

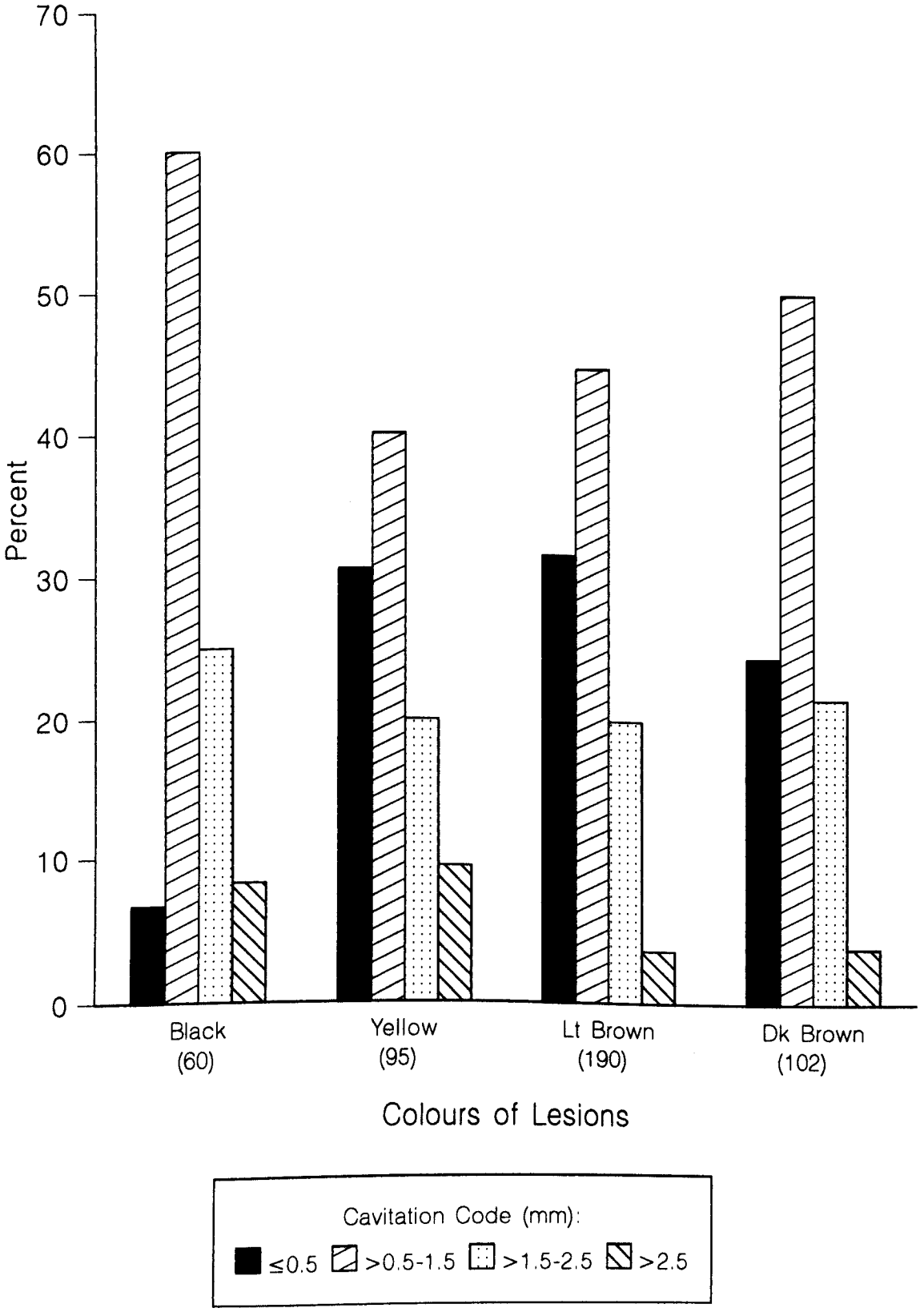


V-212 The Colours of Lesions v Their Locations

The position of the gingival margins of lesions in relation to the crests of the gingival margins themselves are displayed in this histogram. Five categories of Location are defined: 0 mm; >0-1 mm; >1-2 mm; >2-3 mm; and > 3 mm. When the gingival margins of lesions are in the >1-2 mm range of the gingival margin, all Colours of lesions are represented in about the same proportions 28.3 percent; 21.1 percent; 28.9 percent and 30.4 percent respectively from Black to Dark Brown, but amongst lesions at, or very close to the gingival crest, higher proportions of Black and Yellow lesions are found than when they are more distant from the soft tissues. Of lesions at the gingival margin no fewer than 45 percent were Black and 37.9 percent were Yellow, whilst Light Brown and Dark Brown Lesions provided only 20.5 percent and 19.6 percent respectively. In stark contrast only, 1.7 percent of lesions more than 3 mm from the gingival margin were diagnosed as Black and 3.3 percent of those >2-3 mm from it.

The most obvious differences displayed are that the nearer to the gingival margin a lesion is located the more likely it is to be judged Black or Yellow, the further away its Location the more likely it is to be Light or Dark Brown. However, significant proportions of lesions at the gingival margins were either Light Brown (20.5 percent) or Dark Brown (19.6 percent).

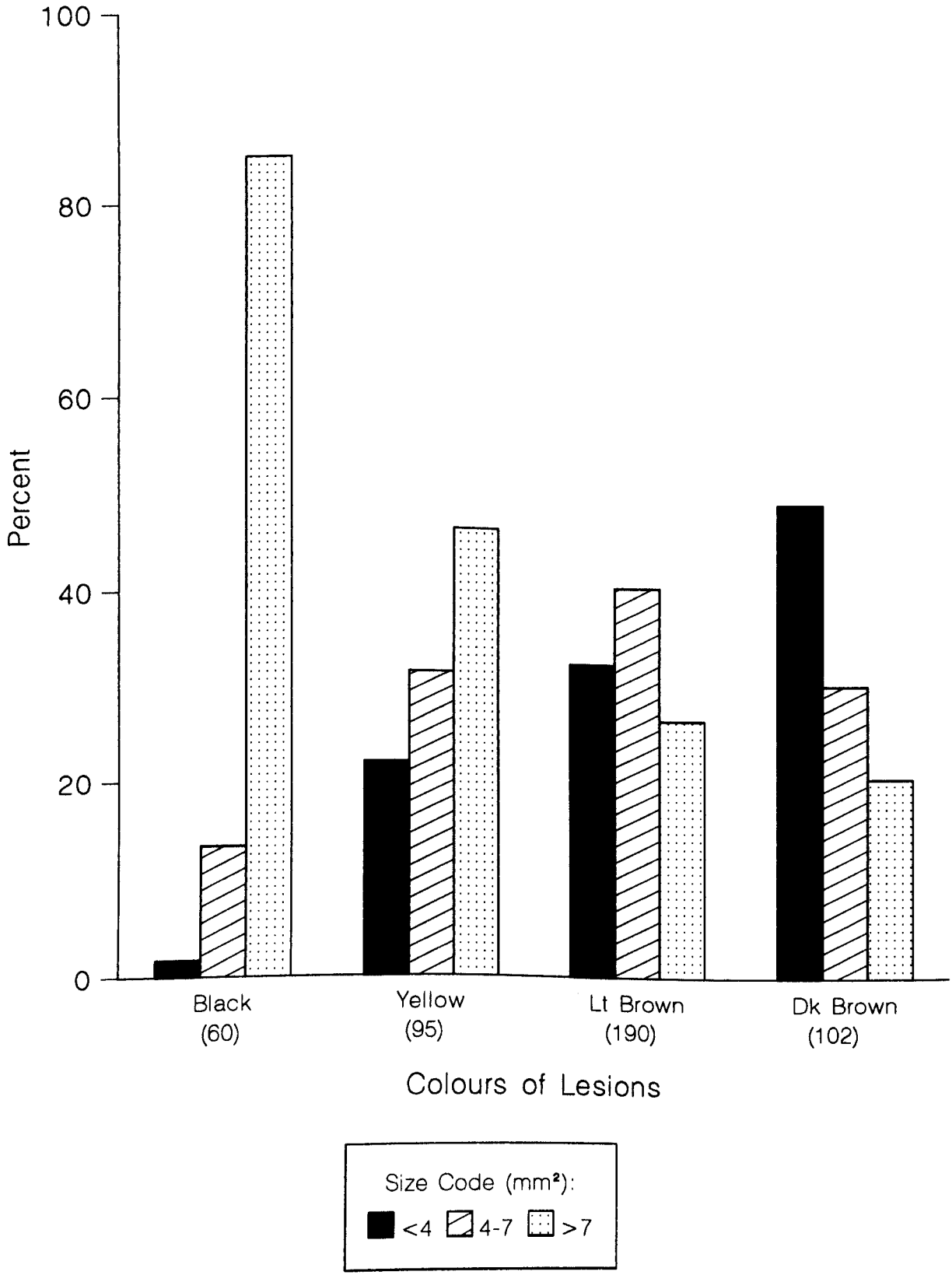
V-213
The Colours of Lesions
V
Their Cavitations



V-213 The Colours of Lesions v Their Cavitations

These data illustrate the estimated losses of root-dentine associated with the four Colours of lesions defined. Four groups of Cavitation depths are shown ie: ≤0.5 mm; >0.5-1.5 mm; >1.5-2.5 mm; and >2.5 mm. Very few non-cavitated lesions were defined as Black (6.7 percent) whilst a very high proportion (60 percent) of shallow lesions (>0.5-1.5 mm) were Black. The other three Colours of carious dentine tend to be more dominant amongst the shallower cavities, reaching maximum proportions in the >0.5-1.5 mm bracket. However, it is clear that amongst the lesions in this study the majority of all types combined were found to be in the >0.5-1.5 mm group (Black: 60 percent; Yellow: 40 percent; Light Brown: 44.7 percent; and Dark Brown: 50 percent) and, perhaps, the most significant observation is that referred to above; the high proportion of Black lesions with a Cavitation of >0.5-1.5 mm. Indeed, the proportions of lesions of various depths which were diagnosed as either Yellow, Light Brown, or Dark Brown are very nearly the same, the exception is found with the Black lesions.

V-214
The Colours of Lesions
V
Their Sizes



V-214 The Colours of Lesions v Their Sizes

The product of the Height and Width of lesions is used as an indicator of their overall Size.

The large Size of Black lesions is very evident for 85 percent are larger than an estimated 7 mm^2 and a high proportion of Yellow lesions (46.3 percent) also fall into this category. Dark and Light Brown lesions are more likely than the others to be small (49 percent of all Dark Brown Lesions were in the <4 mm^2 category).

The mean Size of lesions in each Colour group were also calculated but not presented in a figure. Black lesions were the largest in Size with a mean Size ± standard error (SE) of 15.46 ± 1.27 mm^2 ($P < 0.01$). Yellow lesions with a mean ± SE of 8.88 ± 0.74 mm^2 were also larger than Light Brown or Dark Brown lesions which had Sizes of 6.30 ± 0.36 mm^2 and 4.94 ± 0.43 mm^2 respectively ($P < 0.05$).

V-220 The Textures of Lesions

The relationships between the Textures of lesions and their Colours have been shown in V-211 and, therefore, only three groups of data are presented in this section, ie relating to:

- the Locations of lesions: V-221,
- their Cavitations: V-222, and
- their Sizes: V-223

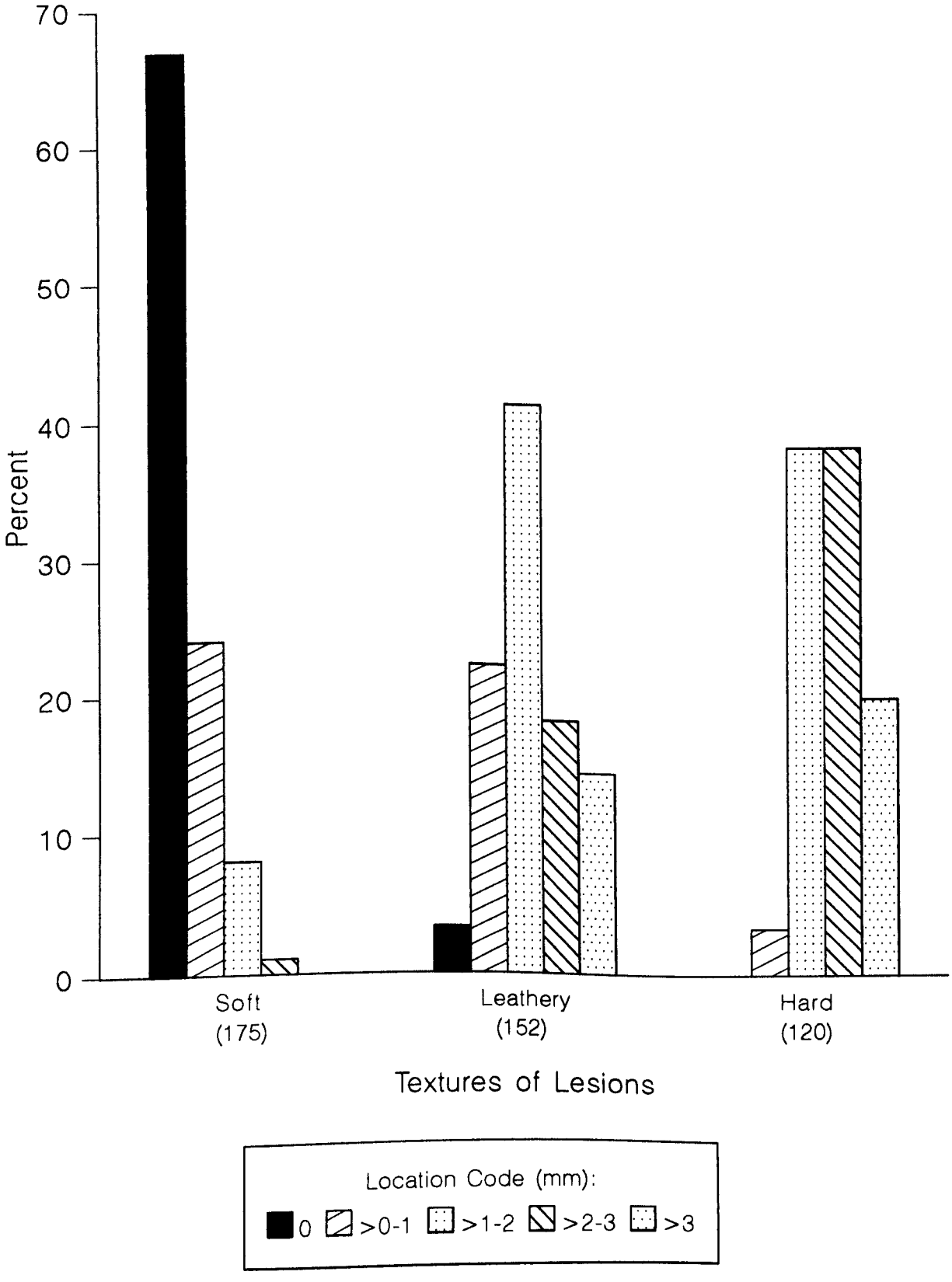
As in V-211 the three categories of Textures are:

- Soft; Leathery; and Hard

The Texture of Lesions in each category were diagnosed as follows:

- 175 Lesions as Soft
- 152 as Leathery and
- 120 as Hard in Texture

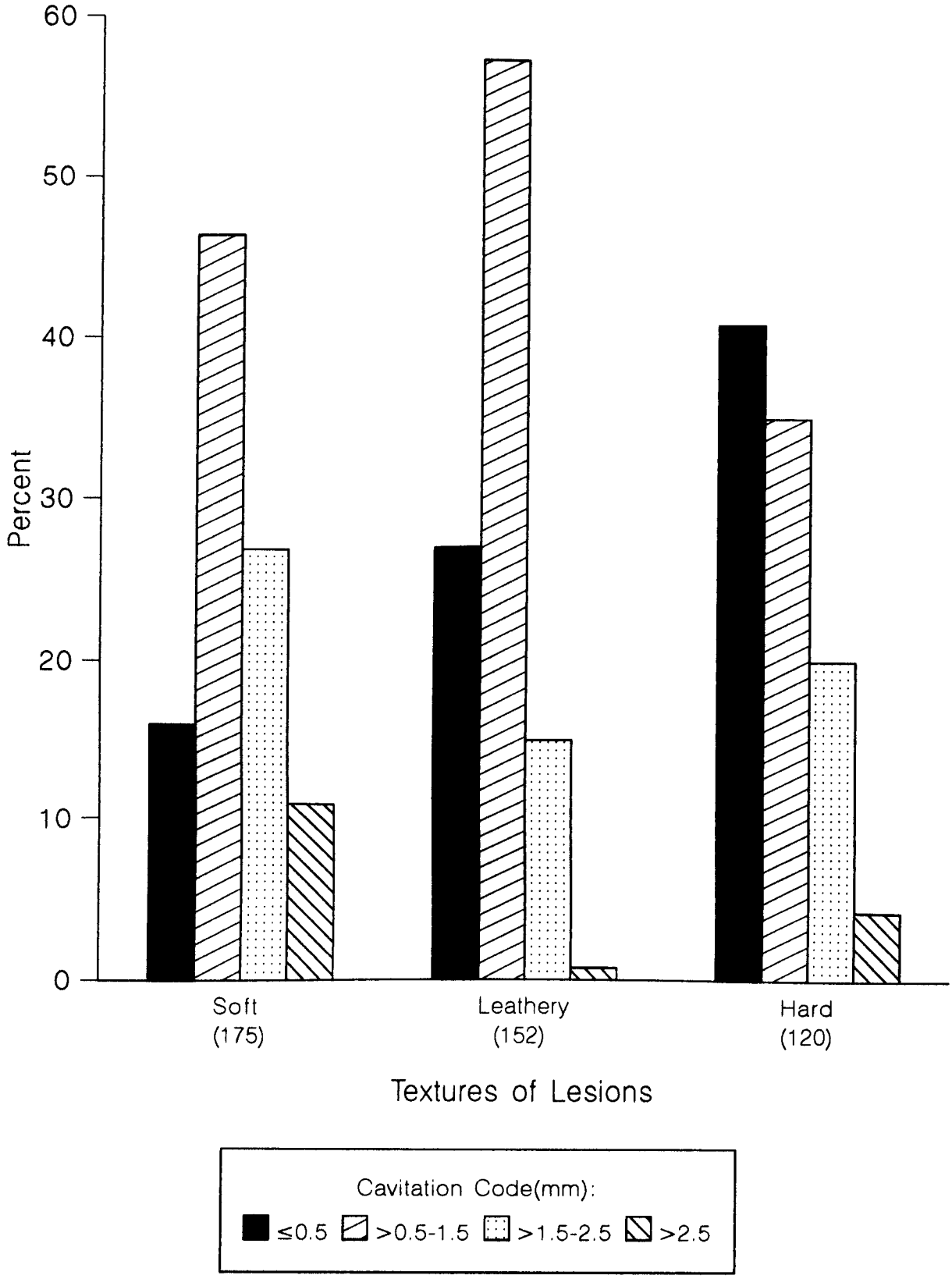
V-221
The Textures of Lesions
V
Their Locations



V-221 The Textures of Lesions v Their Locations

A quite dramatic relationship is revealed by the data between these two variables for, amongst Soft lesions, not less than 66.9 percent were adjacent to the gingival margin and 90.9 percent were 1 mm or less from it. Only 3.3 percent of Leathery lesions and no Hard lesions were found to be located at the gingival margin and only 3.3 percent were 1 mm or less from it. In contrast to this no Soft lesions were found more than 3 mm from the gingival margin and only 1.1 percent were as far as >2-3 mm from it.

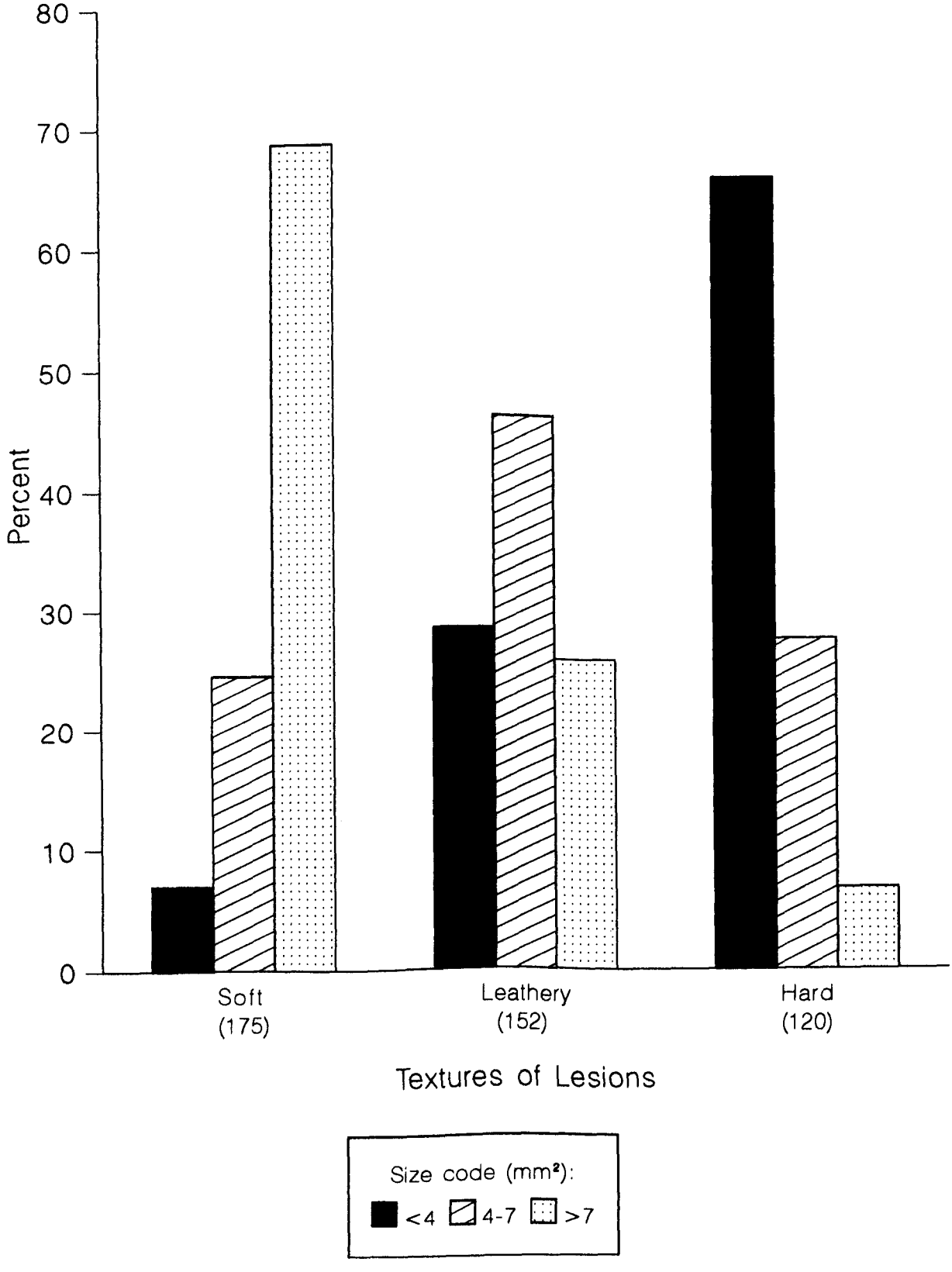
V-222
The Textures of Lesions
V
Their Cavitations



V-222 The Textures of Lesions v Their Cavitations

The proportions of Soft lesions shown in this histogram peak (46.3 percent) with Cavitations in the >0.5-1.5 mm band though 16 percent of such lesions display virtually no Cavitations (≤0.5 mm). Similarly, Leathery lesions predominantly (almost 57.2 percent) showed Cavitations in the >0.5-1.5 mm band, with 27 percent having almost no tissue loss. Hard lesions predominantly fell into the two shallow categories (40.8 percent less than 0.5 mm and 35 percent >0.5-1.5 mm), only 4.2 percent exhibiting Cavitation more than 2.5 mm in depth. The great majority of Leathery lesions were relatively small (27 percent less than 0.5 mm and 57.2 percent being only >0.5-1.5 mm deep) and, with few exceptions, the remainder (15.1 percent) were >1.5-2.5 mm deep.

V-223
The Textures of Lesions
V
Their Sizes



V-223 The Textures of Lesions v Their Sizes

The product of the Height and Width of lesions is used as an indicator of their overall Size.

These data clearly demonstrate that Soft lesions tend to be large whilst Hard lesions tend to be small. 68.6 percent of all Soft lesions estimated to be >7 mm^2 whilst only 6.9 percent were <4 mm^2, and 24.6 percent were 4-7 mm^2. Almost the opposite distribution was found amongst the Hard lesions (65.8 percent: <4 mm^2; 27.5 percent: 4-7 mm^2; and only 6.7 percent more than 7 mm^2. Leathery lesions were found in much nearer proportions in the three Sizes (28.3 percent: <4 mm^2; 46.1 percent: 4-7 mm^2 and 25.7 percent more than 7 mm^2).

The mean Size of lesions in each Texture group were also calculated but were not presented in a figure. Soft lesions were the largest in Size with a mean Size ± SE of 12.19 ± 0.63 mm^2, whilst Leathery lesions, with a mean Size ± SE of 6.28 ± 0.38 mm^2, were larger than Hard Primary Root Caries with a mean Size ± SE of 3.21 ± 0.19 mm^2. These differences show statistical significance at probability of 0.01.

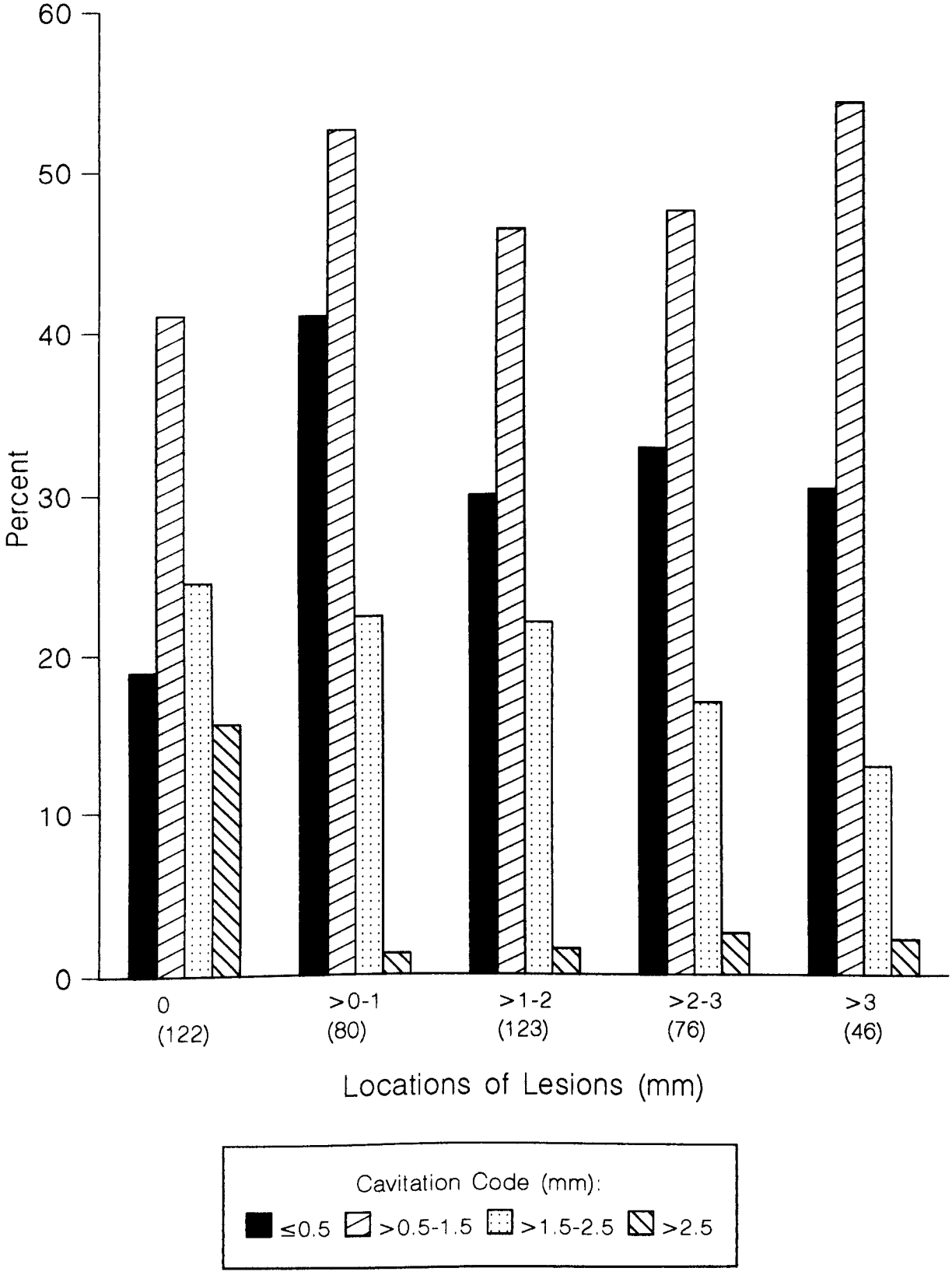
V-230 Locations of Lesions

Two histograms represent the data relating to the Locations of Lesions to their other clinical characteristics which have not already been presented in respect of their Colour (V-212) and their Texture (V-221), ie The Locations of Lesions v Their Cavitations (V-231) and their Sizes (V-232).

Location has been defined (IV-311) as the minimum distance from the gingival margin of the lesion and the crest of the gingivae itself.

- 122 Lesions were diagnosed to be at the gingival margin;
- 80 were less than 1 mm above it;
- 123 between >1 and 2 mm;
- 76 between >2 and 3 mm; and
- 46 were more than 3 mm from the gingival margin

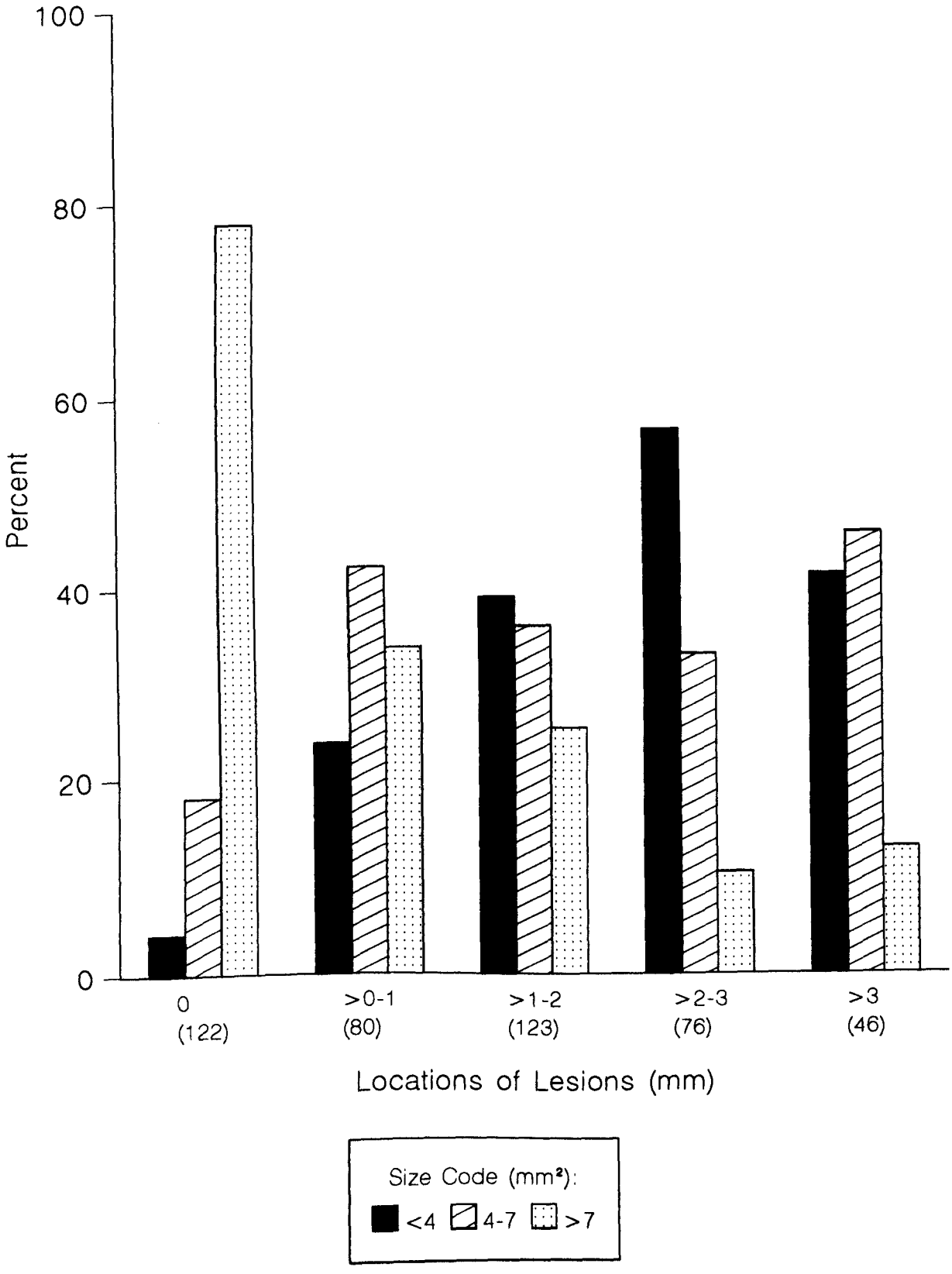
V-231
The Locations of Lesions
V
Their Cavitations



V-231 The Locations of Lesions v Their Cavitations

The highest proportions of all lesions were found to have Cavitations in the >0.5-1.5 mm range of depths (41 percent; 52.5 percent; 46.3 percent; 47.4 percent and 54.3 percent respectively) and, with the exception of those lesions at the gingival margin, all the other Locations of lesions revealed the second highest proportions to be in the smallest Cavitation group of ≤0.5 mm (18.9 percent; 41 percent; 30.1 percent; 32.9 percent; and 30.4 percent respectively). What seems to be the most dramatic finding is that whilst 15.6 percent of lesions located at the gingival margin were more than 2.5 mm deep, only 1.3 percent; 1.6 percent; 2.6 percent and 2.2 percent of lesions in the >0-1 mm, >1-2 mm, >2-3 mm and over 3 mm categories, respectively were diagnosed.

V-232
The Locations of Lesions
V
Their Sizes



V-232 The Locations of Lesions v Their Sizes

The product of the Height and Width of lesions is used as an indicator of their overall Size.

This histogram clearly reveals that the great majority (77.9 percent) of lesions found to be located at the gingival margin were large, in excess of 7 mm^2 and, in general, the smaller lesions (<4 mm^2) formed increasing proportions the further from the gingivae they were located, ie 4.1 percent at the margin; 23.8 percent >0-1 mm from it; 39 percent: >1-2 mm; 56.6 percent: >2-3 mm from it, falling back to 41.3 percent when more than 3 mm from the margin. However, the two larger sized groups together formed 87 percent of all lesions most remote from the gingivae.

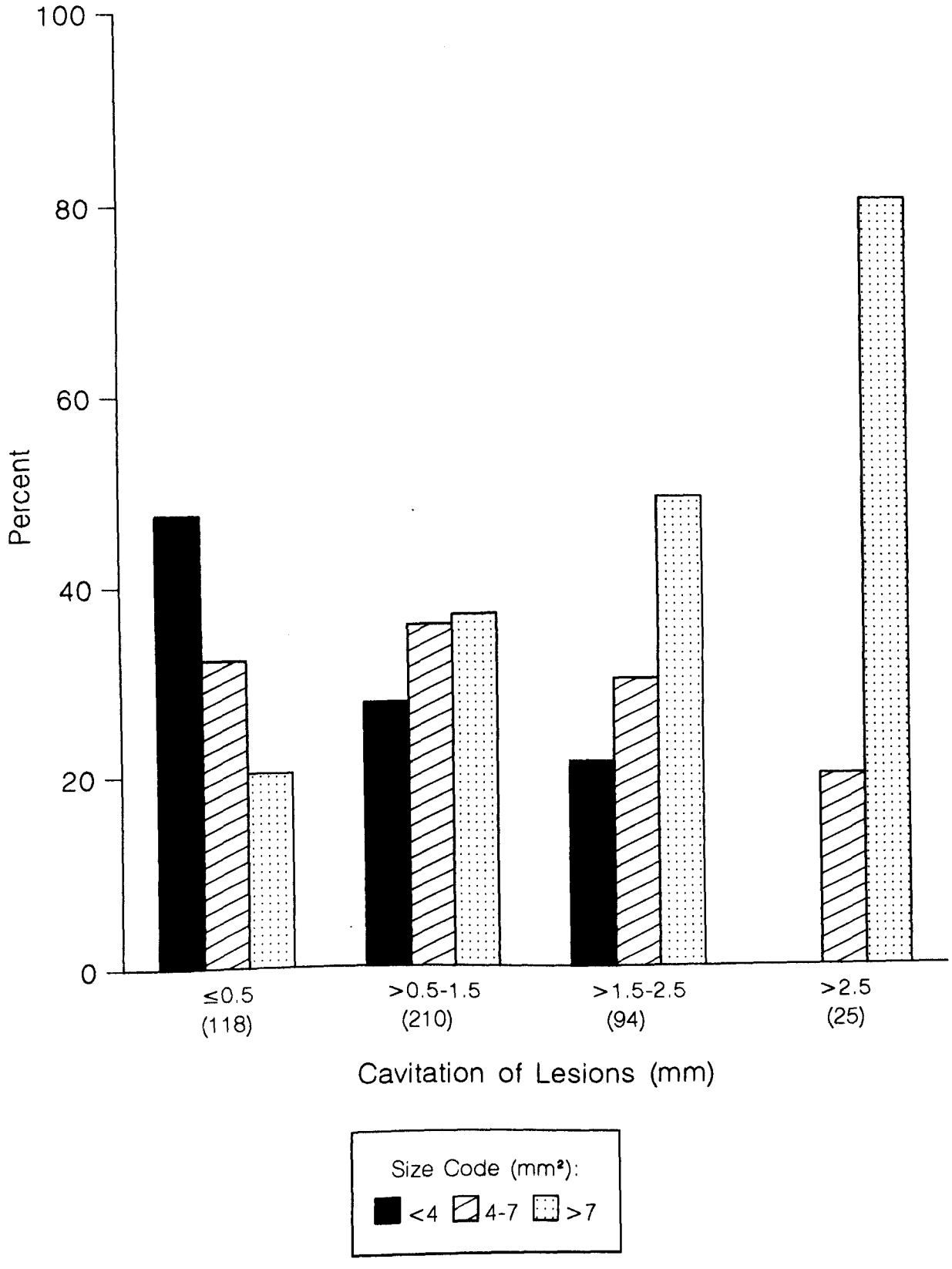
The mean Size of lesions in each Location group were also calculated but not presented in a figure. Lesions located at the gingival margin with a mean Size ± SE of 13.41 ± 0.77 mm^2, were significantly larger in Size than other Location groups and this significance between groups was determined by using $P < 0.01$. Lesions located >0-1 mm from the gingival margin had a mean Size ± SE of 7.25 ± 0.63 mm^2, which were larger than lesions located >2-3 and >3 mm from it with mean Sizes ± SE of 4.13 ± 0.37 mm^2 and 4.43 ± 0.39 mm^2 respectively ($P < 0.05$). Lesions located >1-2 mm from the gingival margin with a mean Size of 6.02 ± 0.52 mm^2, were in turn larger than lesions >3 mm from the gingiva ($P < 0.05$).

V-240 The Cavitations of Lesions

Only one group of data needs to be presented under this heading: The Cavitations of Lesions v Their Sizes (V-241) for the others have already been given. Cavitation has been defined (IV-311) as the greatest loss of surface contour as measured by recording the greatest distance between the existing surface of the lesion and what was judged to have been the original root surface. The four groupings of the Primary Root Caries lesions in this section are:

- 0.5 mm or less Cavitation:- 118 lesions;
- >0.5 to 1.5 mm Cavitation:- 210 lesions;
- >1.5 to 2.5 mm Cavitation:- 94 lesions; and
- more than 2.5 mm Cavitation:- 25 lesions

V-241
The Cavitations of Lesions
V
Their Sizes



V-241 The Cavitations of Lesions v Their Sizes

This clearly shows how lesions with large Sizes are most likely to have more loss of surface contour than lesions with smaller Sizes. No lesions with Sizes less than 4 mm^2 were found with Cavitation in excess of 2.5 mm whilst some 80 percent of lesions with such Cavitations had Sizes in excess of 7 mm^2. Thus, the lesions with more loss of surface contour tend to be large in Size.

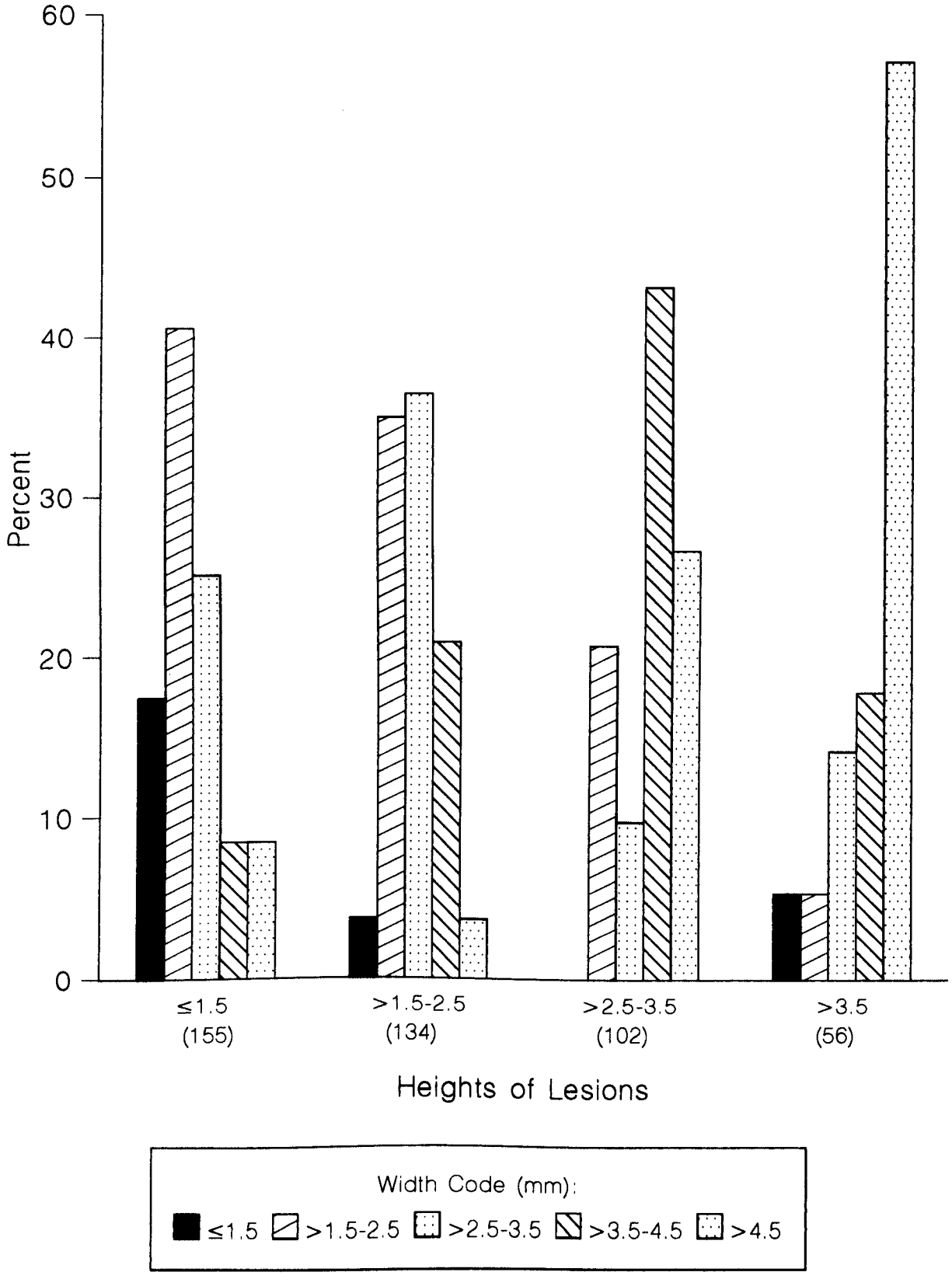
The mean Size of lesions in each Cavitation group were also calculated. Lesions with Cavitation >2.5 mm had a mean Size ± SE of 16.56 ± 2.47 mm^2 which was significantly greater than the Size of the other Cavitation groups ($P < 0.01$). Primary Root Caries lesions with Cavitation of >1.5-2.5 and >0.5-1.5 had mean ± SE Sizes of 8.99 ± 0.68 mm^2 and 7.59 ± 0.45 mm^2 respectively and these were significantly greater in Size than lesions with a Cavitation of ≤0.5 mm which had a mean ± SE Size of 5.27 ± 0.45 mm^2. The significance level in this latter analysis was set at $P < 0.05$.

V-250 Heights and Widths of Lesions

Since the Heights and Widths of lesions, together with the products of these dimensions on which the estimate of area is made are each ways of assessing the Size of lesions these three dimensions are not presented separately in relation to the other clinical signs of Primary Root Caries. However, it is of interest to record the relationship between Heights and Widths in order to assess the general shapes these lesions present, whether high and narrow or low and wide.

Height and Width of lesions has been defined (IV-311) as the maximum occluso-gingival and the maximum mesio-distal or bucco-lingual dimensions.

V-251
The Heights of Lesions
V
Their Widths



V-251 The Heights of Lesions v Their Widths

Perhaps, not surprisingly, lesions in excess of 3.5 mm High are also likely to be very Wide, 57.1 percent of these being more than 4.5 mm wide, in contrast to only 10.8 percent being less than 2.5 mm wide. Lesions of 1.5 mm or less in Height tend to be narrow (17.4 percent 1.5 mm or less ; 40.6 percent between >1.5 and 2.5 mm wide; and 25.2 percent between >2.5 and 3.5 mm). Also lesions were wider than they were high.

V-300 The Clinical Signs of Lesions and Their Perceived Treatment Needs

It is important to emphasise that the Perceived Treatment Needs were defined by the author with respect to each Primary Root Caries Lesion included in the study and are, as a result, the consequence of his own judgements and opinions. They cannot, therefore, be presumed to relate in any direct way to the Perceived Treatment Needs that would be found in a random study of numerous dentists' prescriptions.

The four categories of Perceived Treatment Needs have been defined and their relationships to the Clinical Signs will be presented in five histograms as follows:

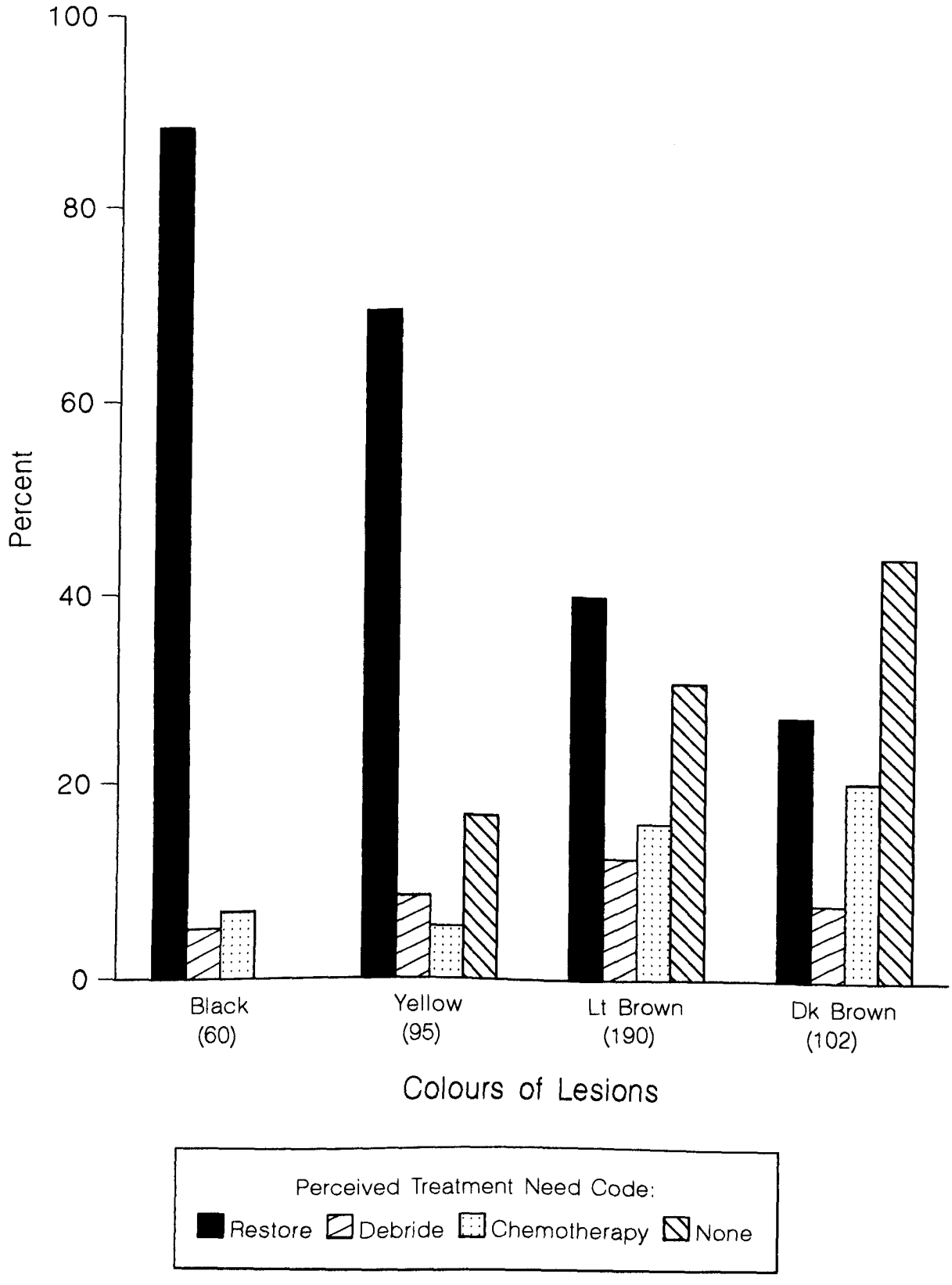
- The Colours of Lesions v Their Perceived Treatment Needs : V-301
- The Textures of Lesions v Their Perceived Treatment Needs : V-302
- The Locations of Lesions v Their Perceived Treatment Needs : V-303
- The Cavitations of Lesions v Their Perceived Treatment Needs : V-304
- The Sizes of Lesions v Their Perceived Treatment Needs : V-305

As described in IV-312 Perceived Treatment Needs were defined as:

- None [120];
- Chemotherapy [61];
- Carious Dentine Removed (Debride) [43]; or
- Caries Removed and the Lesion Restored [223].

The numbers of lesions in each category are shown in parenthesis.

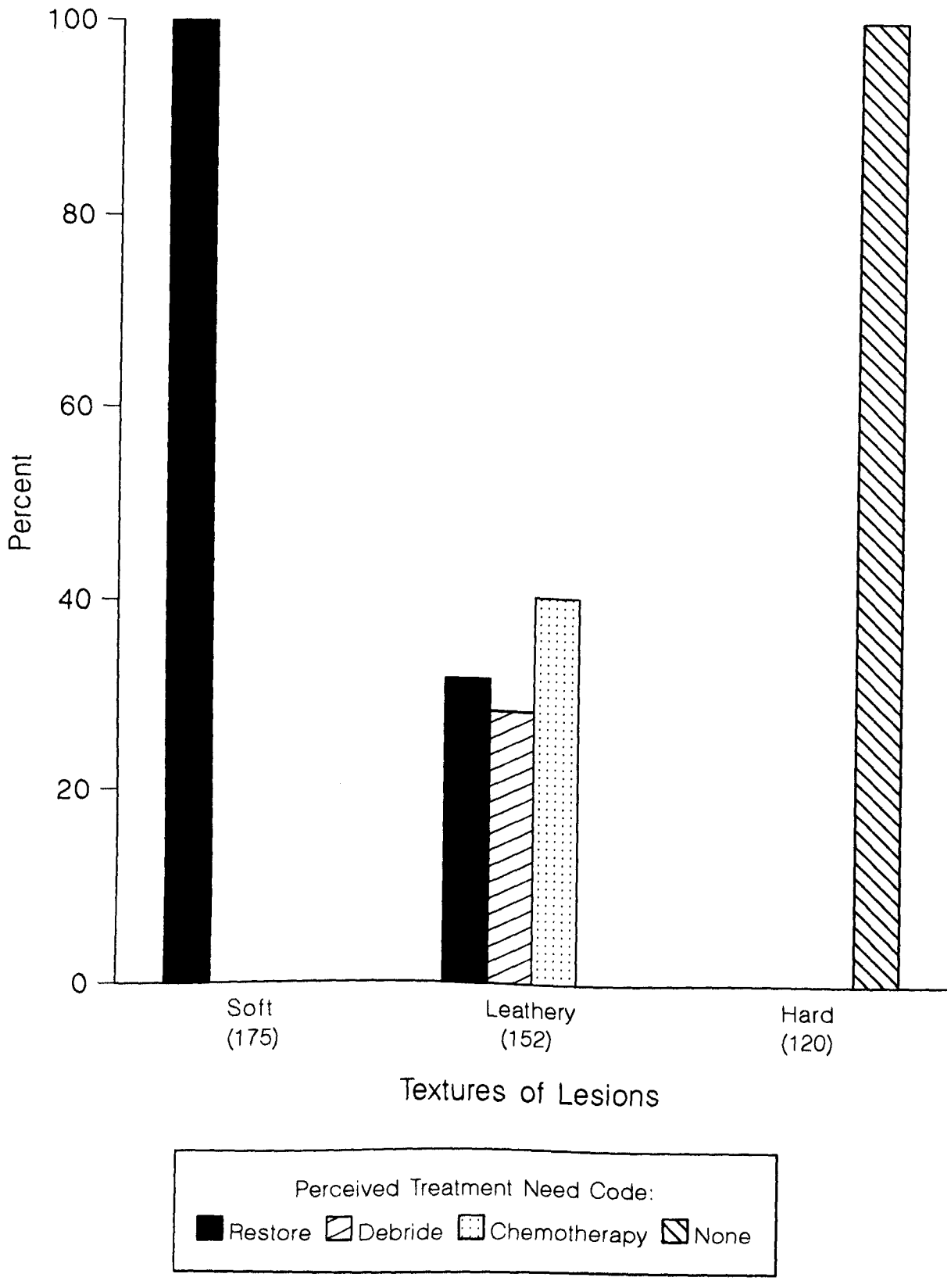
V-301
The Colours Of Lesions
V
Their Perceived Treatment Needs



V-301 The Colours of Lesions v Their Perceived Treatment Needs

This histogram clearly shows the ranking of lesions by Colour and their clinical management as prescribed by the author, ie Black : Yellow : Light Brown: Dark Brown. No fewer than 88.3 percent of all Black lesions were restored, a total of only 11.7 percent being managed by any other strategy. 69.5 percent of Yellow lesions; 40 percent of Light Brown and 27.5 percent of Dark Brown lesions were also restored. In contrast, 44.1 percent of Dark Brown lesions; 31.1 percent of Light Brown; 16.8 percent of Yellow; but no Black lesions were diagnosed as not requiring treatment of any kind. The removal of carious dentine only, without other intervention was relatively rarely prescribed (Black: 5 percent; Yellow: 8.4 percent; Light Brown: 12.6 percent and Dark Brown: 20.6 percent). The figures in the Chemotherapy groups (Black: 6.7 percent; Yellow: 5.3 percent; Light Brown: 16.3 percent; and Dark Brown: 20.6 percent) relate to lesions which would otherwise have been restored.

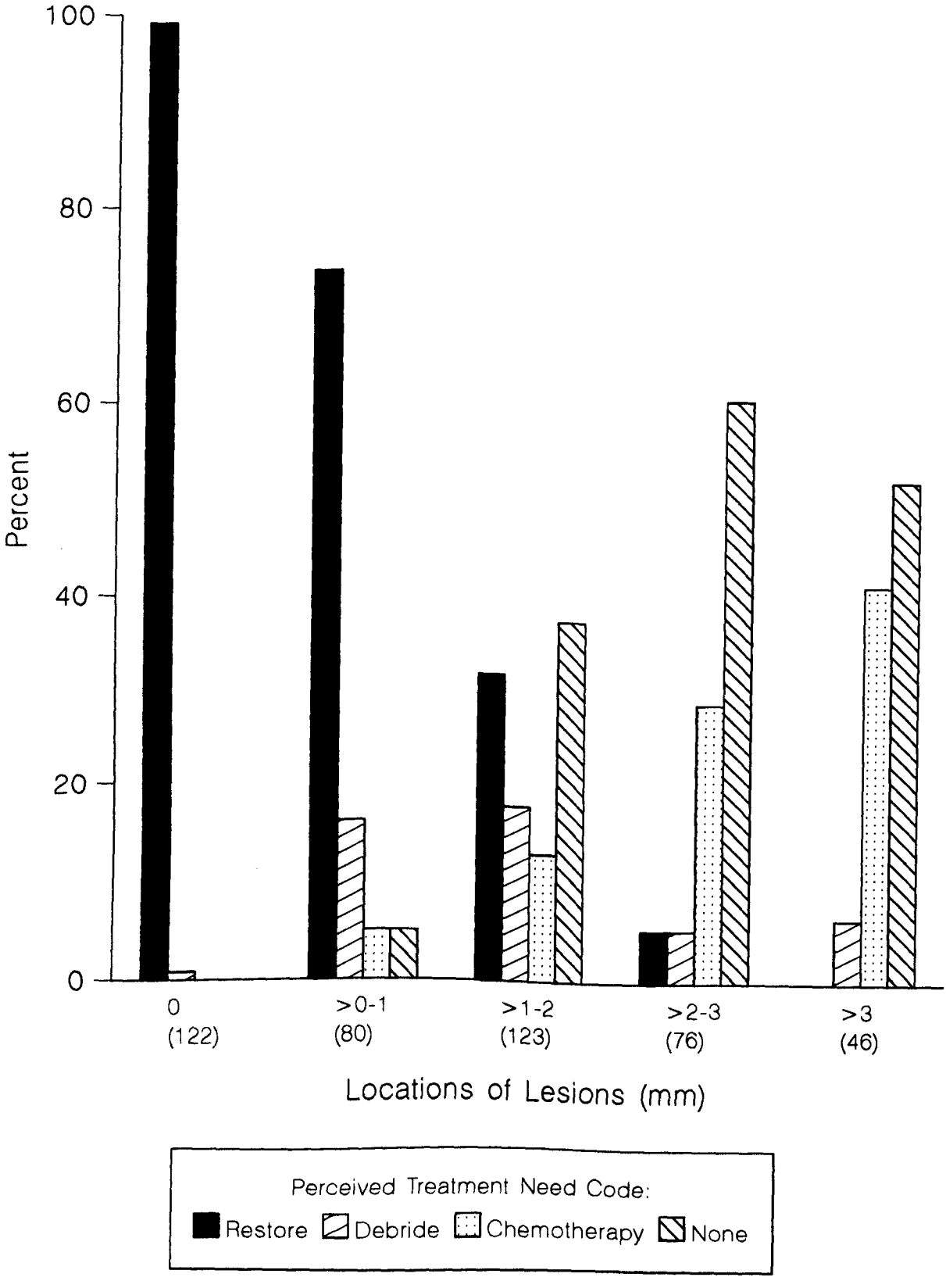
V-302
The Textures of Lesions
V
Their Perceived Treatment Needs



V-302 The Textures of Lesions v Their Perceived Treatment Needs

These figures quite clearly show the major factor on which the author chose to base the treatment strategy, ie all Soft Carious lesions were Restored; all Hard lesions were not treated; whilst the Leathery lesions were subjected in almost equal proportions to either Restoration, Debridement, or Chemotherapy but none were left untreated. Indeed, it is necessary to recall that lesions selected for Chemotherapy were identified only from the Leathery-textured group.

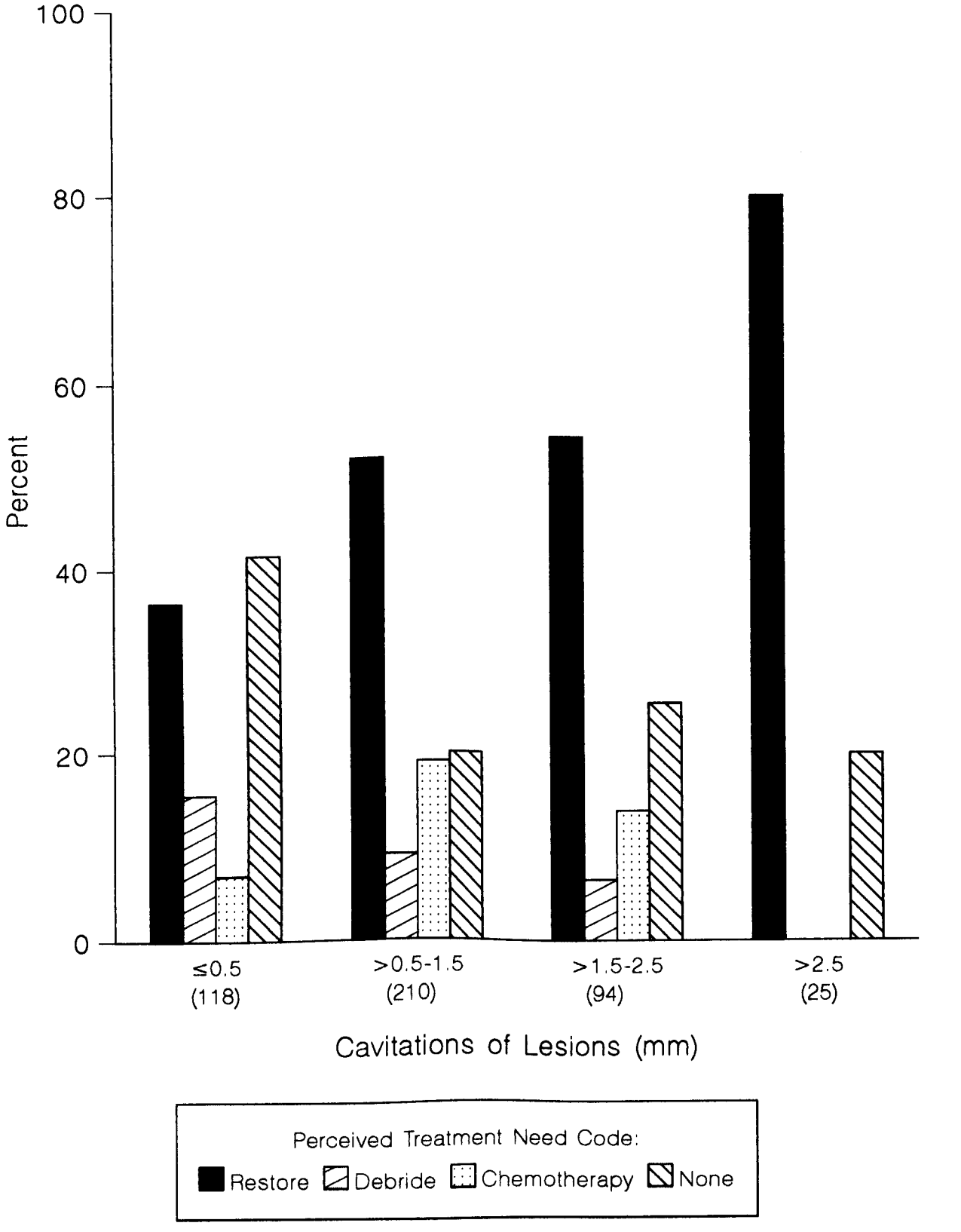
V-303
The Locations of Lesions
V
Their Perceived Treatment Needs



V-303 The Locations of Lesions v Their Perceived Treatment Needs

These data clearly show that almost every lesion (99.2 percent) located at the gingival margin was deemed to require a restoration, whilst the proportions of lesions restored in the other four Locations, progressively further from the gingival margin, were 73.8 percent; 31.7 percent; 5.3 percent and 0 percent respectively. In contrast no treatment was prescribed for 5 percent located: >0-1 mm; 37 percent located: >1-2 mm; 60.5 percent located: >2-3 mm and 52.2 percent located more than 3 mm from the gingival crest. Those selected for Chemotherapy formed higher proportions of lesions located some distance from the gingival margin (>1-2 mm: 13 percent; >2-3 mm: 28.9 percent; and more than 3 mm: 41.3 percent) whilst none were selected from the group located at the gingival margin and only 5 percent of those at >0-1 mm were selected.

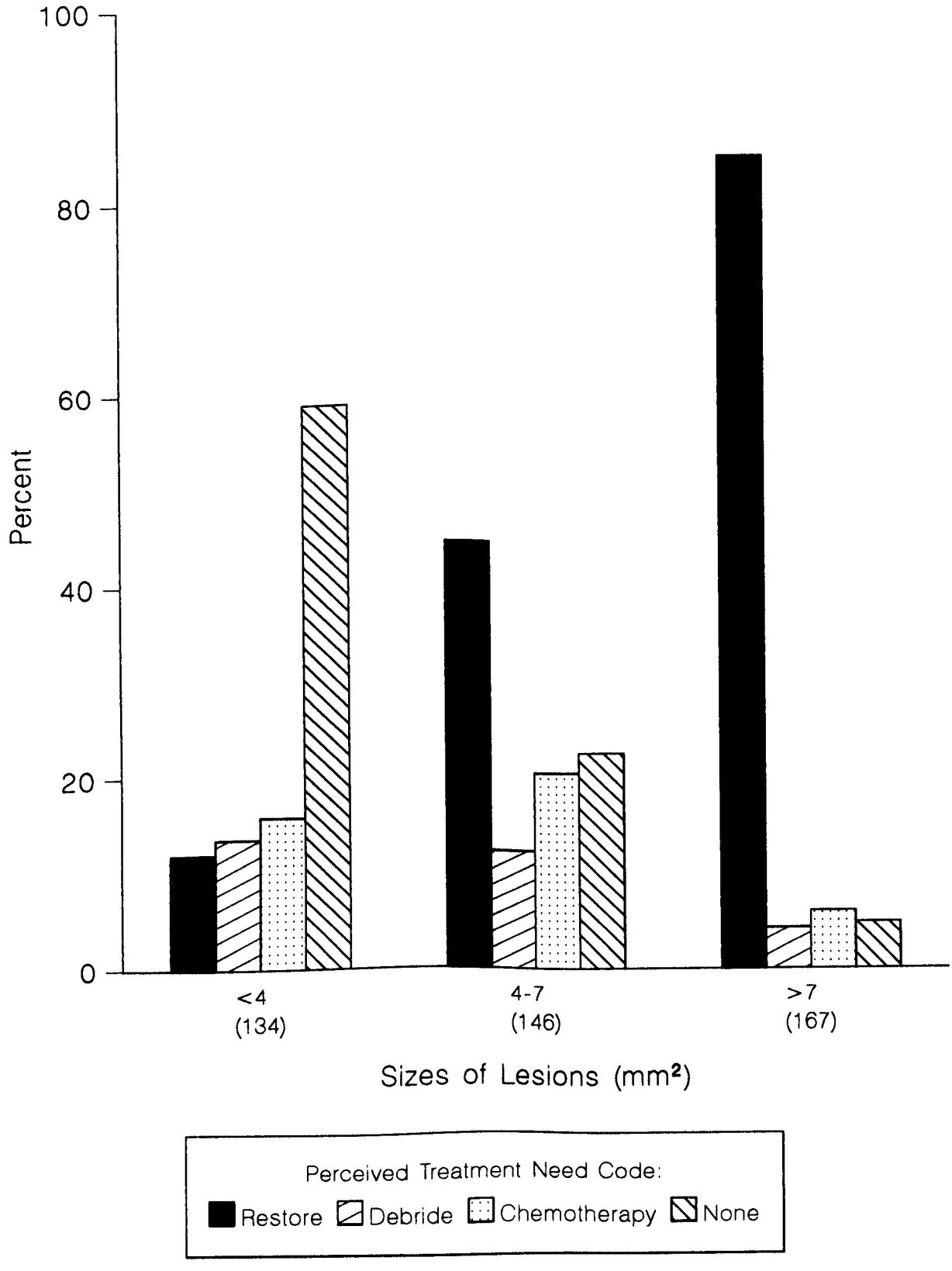
V-304
The Cavitations of Lesions
V
Their Perceived Treatment Needs



V-304 The Cavitations of Lesions v Their Perceived Treatment Needs

Increasingly higher proportions of lesions were prescribed restorative management in the groups with greater Cavitation of the dentine. 80 percent of all lesions cavitated to depths in excess of 3 mm were restored, the remainder were not considered to need any treatment at all. Of the other Cavitation groups ≤0.5 mm: 36.4 percent; >0.5-1.5 mm: 51.9 percent; >1.5-2.5: 54.3 percent were restored, whilst the opposite order of frequency prevailed with respect to no treatment (41.5 percent; 20 percent; 25.5 percent and 20 percent, respectively) ie higher proportions of small lesions rather than larger lesions went untreated.

V-305
The Sizes of Lesions
V
Their Perceived Treatment Needs



V-305 The Sizes of Lesions v Their Perceived Treatment Needs

The product of the Height and Width of lesions is used as an indicator of their overall Size.

Here it is revealed that the larger the Size of a lesion the greater the likelihood of it being restored, whilst the smaller the lesions the more likely it is that no treatment will be prescribed, ie of the larger sized group of more than 7 mm^2, 85 percent were restored; whilst 59 percent of the smallest lesions less than 4 mm^2, went untreated.

The mean Size of lesions in each Perceived Treatment Need group was also calculated but was not presented in a figure. Lesions deemed to require a restoration with a mean Size ± SE of 11.43 ± 0.53 mm^2 were significantly larger than any of the other groups. Lesions deemed to require Chemotherapy with a mean Size ± SE of 5.27 ± 0.46 mm^2 were significantly larger in Size than lesions deemed to require no treatment with a mean Size ± SE of 3.21 ± 0.19 mm^2 ($P < 0.05$). Lesions deemed to require debridement only had a mean Size ± SE of 5.05 ± 0.63 mm^2 which was not significantly different in Size from the latter two mentioned groups.

V-400 The Clinical Signs of Lesions Related to the Microbiology of Carious Dentine

The Microbiology of Carious Primary Root Dentine is presented with respect to the four groups of organisms discussed in Section IV ie Gram-positive pleomorphic rods (GPPR); Mutans streptococci; Lactobacilli; and Yeasts; in addition to the total numbers of micro-organisms recovered (total colony forming units or CFUs) are also presented. Different pictures are revealed according to the ways in which the data on the microbiological picture are presented relevant to each of the seven clinical signs, as previously reported. These are:

- The Numbers of Dentine Micro-organisms;
- The Proportions of Dentine Micro-organisms; and
- The Frequency of Isolation of Dentine Micro-organisms

The five groups of Clinical Signs are:

- The Colours of Lesions : V-410
- The Textures of Lesions : V-420
- The Locations of Lesions : V-430
- The Cavitations of Lesions : V-440
- The Sizes of Lesions : V-450

V-410 The Colours of Lesions and Their Microbiology

The four Colours previously defined were identified and the lesions placed in these categories ie Black, Yellow, Light Brown, and Dark Brown.

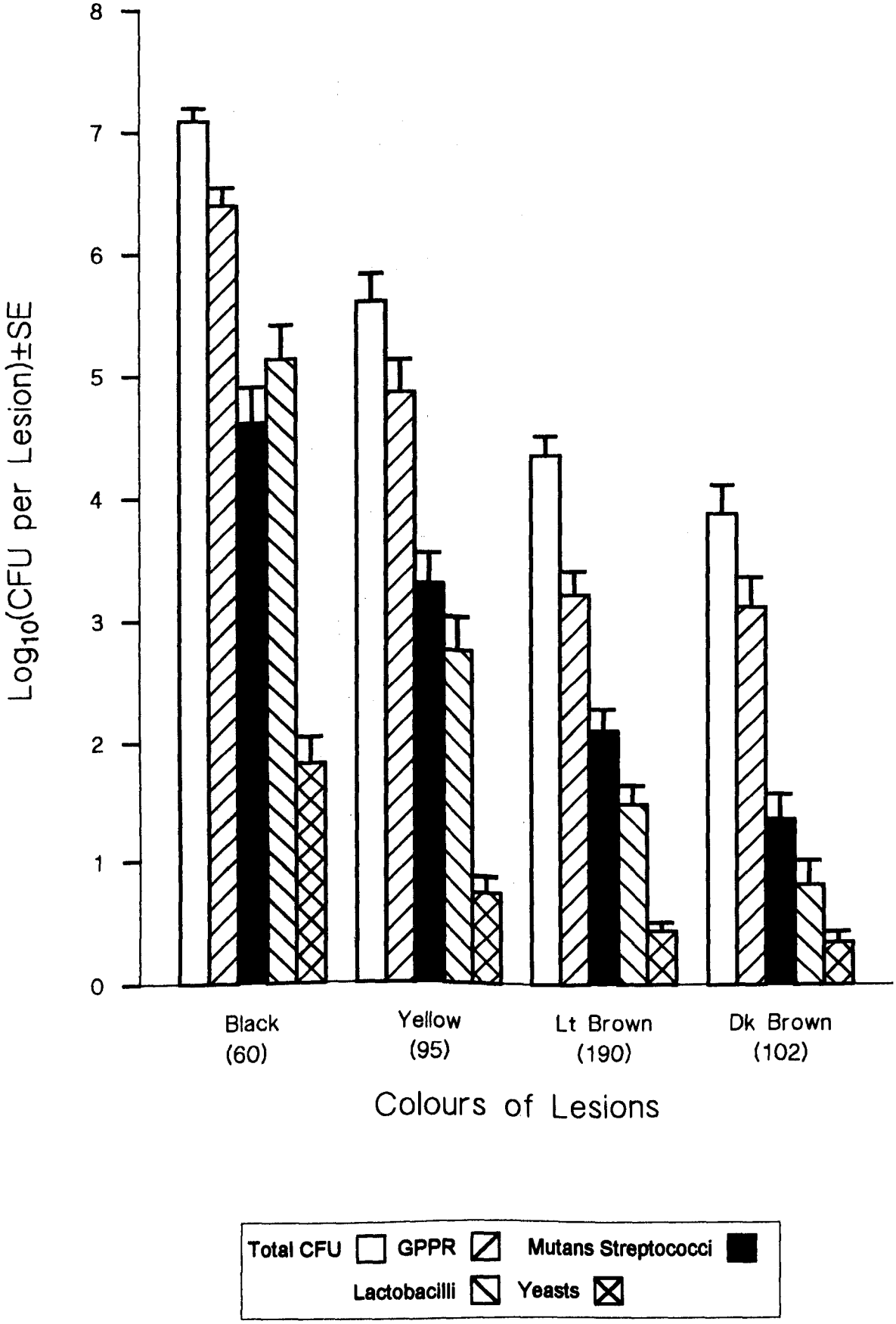
The 447 lesions studied were defined as follows:

- 60 as Black;
- 95 as Yellow;
- 190 as Light Brown and
- 102 as Dark Brown

The total numbers of Colony forming units in each of these categories and the constituent four types of micro-organisms are presented as follows:

- The Colours of Lesions v The Numbers of Dentine Micro-organisms : V-411
- The Colours of Lesions v The Proportions of Dentine Micro-organisms : V-412
- The Colours of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-413

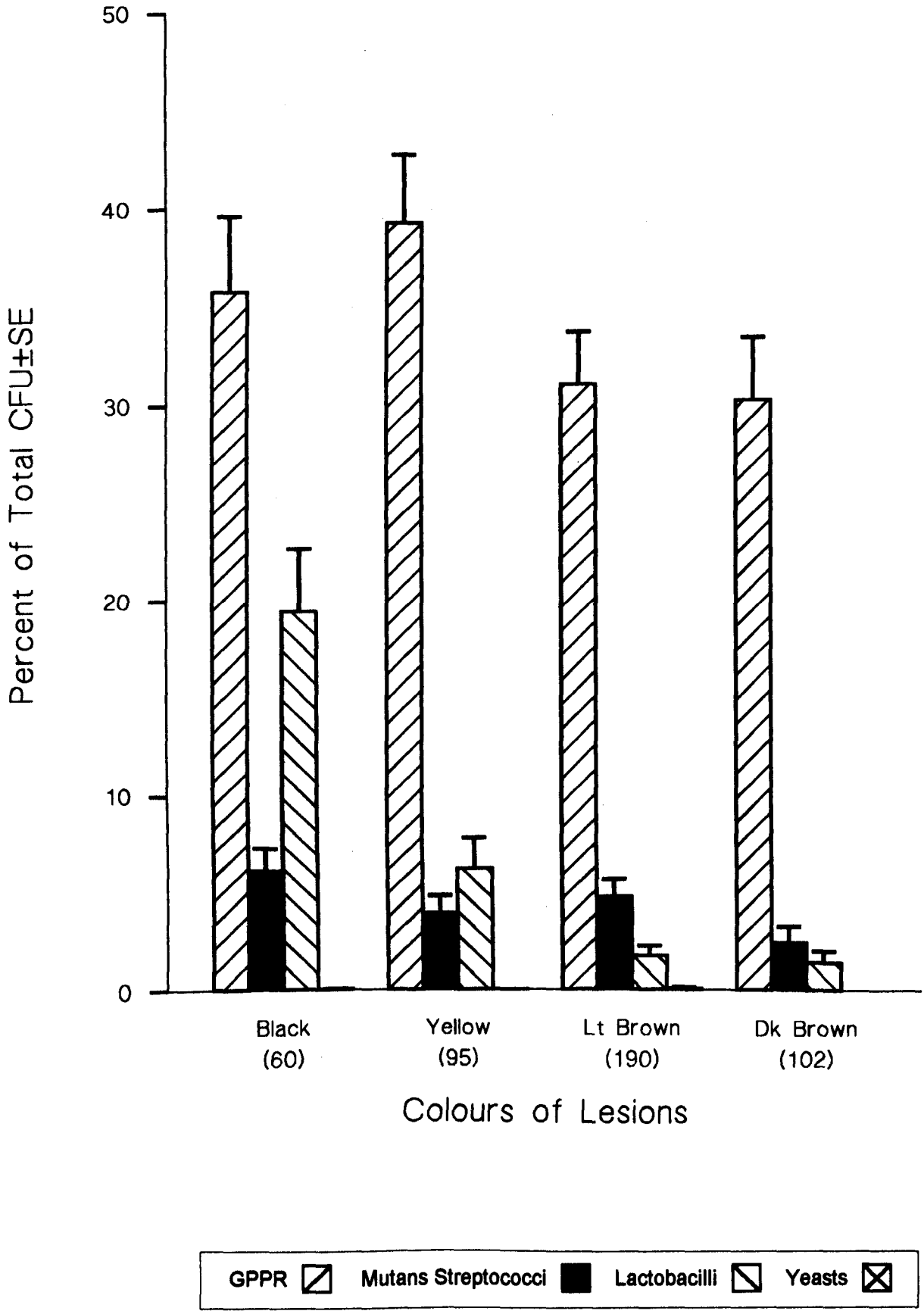
V-411
The Colours of Lesions
V
The Numbers of Dentine Micro-organisms



V-411 The Colours of Lesions v The Numbers of Dentine Micro-Organisms

These data reveal quite clearly that the total numbers of Colony forming units that could be cultured from Primary Root Caries Lesions could rank the lesions by Colour, the highest numbers being found in Black Lesions. The differences are dramatic for Black Lesions contained a mean ± standard error of Log_{10} 7.09 ± 0.11 which is significantly greater than Yellow Lesions with Log_{10} 5.59 ± 0.23 which in turn is also significantly greater than Light Brown Lesions with Log_{10} 4.34 ± 0.16 and Dark Brown Lesions with only Log_{10} 3.87 ± 0.23 ($P < 0.01$). Though the four specific taxa of micro-organisms do not all follow precisely the same pattern, the overall picture is the same, the most notable exception being the relatively high numbers of Lactobacilli found in the Black Lesions compared with the other Colours of carious dentine ($P < 0.001$).

V-412
The Colours of Lesions
V
The Proportions of Dentine Micro-organisms



V-412 The Colours of Lesions v The Proportions of Dentine Micro-Organisms

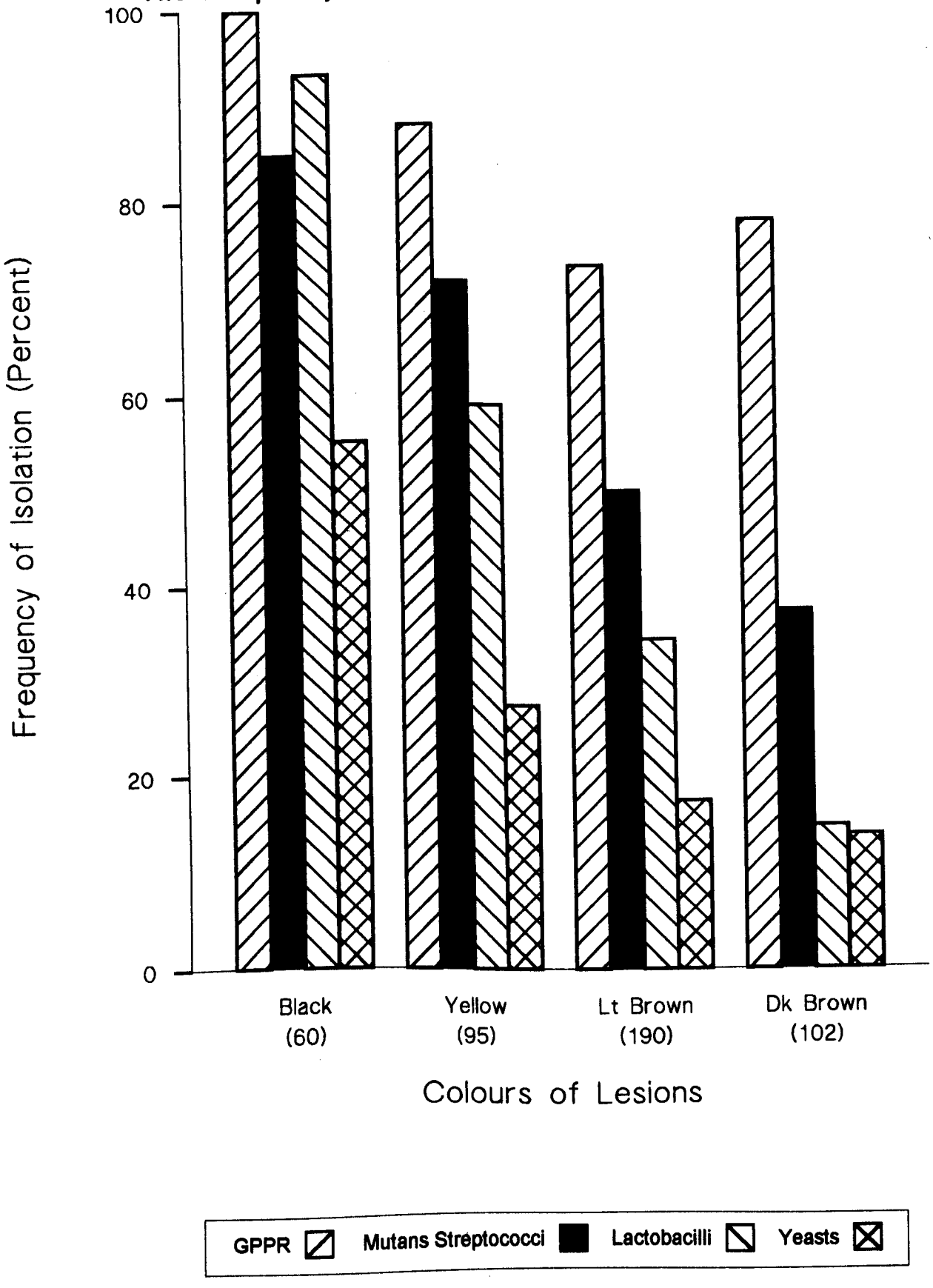
The dominant micro-organisms found in all Colours of carious dentine are clearly the Gram-positive pleomorphic rods, for they form between 31 percent and 39 percent which were not significantly different from one another. The major difference between the four Colours of carious dentine is even more obviously evident in this histogram than in V-411, ie the 19.5 percent of Lactobacilli in the Black lesions which was significantly greater than the 6.3 in Yellow which in turn was significantly greater than either the 1.7 percent in Light Brown or the 1.3 percent in Dark Brown Lesions ($P < 0.05$). The proportions of Yeasts found were minute and, though the Mutans streptococci formed a significantly higher proportion of micro-organisms in Black lesions (6.2 percent) than in the other Colours of lesions (Yellow: 4.0 percent; Light Brown: 4.8 percent and Dark Brown 2.4 percent) the differences were not as great as with the Lactobacilli counts ($P < 0.05$).

V-413

The Colours of Lesions

V

The Frequency of Isolation of Dentine Micro-organisms



V-413 The Colours of Lesions v The Frequency of Isolation of Dentine Micro-organisms

Gram-positive pleomorphic rods were isolated from every single Black lesion and indeed from the vast majority of all the other Colours of lesions (Yellow: 88 percent; Light Brown: 73 percent and Dark Brown : 78 percent). These differences were not significant at the 0.05 probability level. 85 percent of all Black Lesions contained Mutans streptococci and 93 percent, Lactobacilli, but only 55 percent contained any Yeasts. The percentage of Yellow, Light Brown and Dark Brown lesions producing Mutans streptococci, Lactobacilli and Yeasts respectively were progressively fewer from one Colour to the next.

V-420 The Textures of Lesions and Their Microbiology

The standard three Textures of lesions : Soft; Leathery and Hard are once again shown in these data.

Of the 447 lesions the Textures were judged to be as follows:

- 175 Soft;
- 152 Leathery
- 120 Hard

As in V-410 the total numbers of colony forming units and of Gram-positive pleomorphic rods, Mutans streptococci, Lactobacilli and Yeasts are presented as follows:

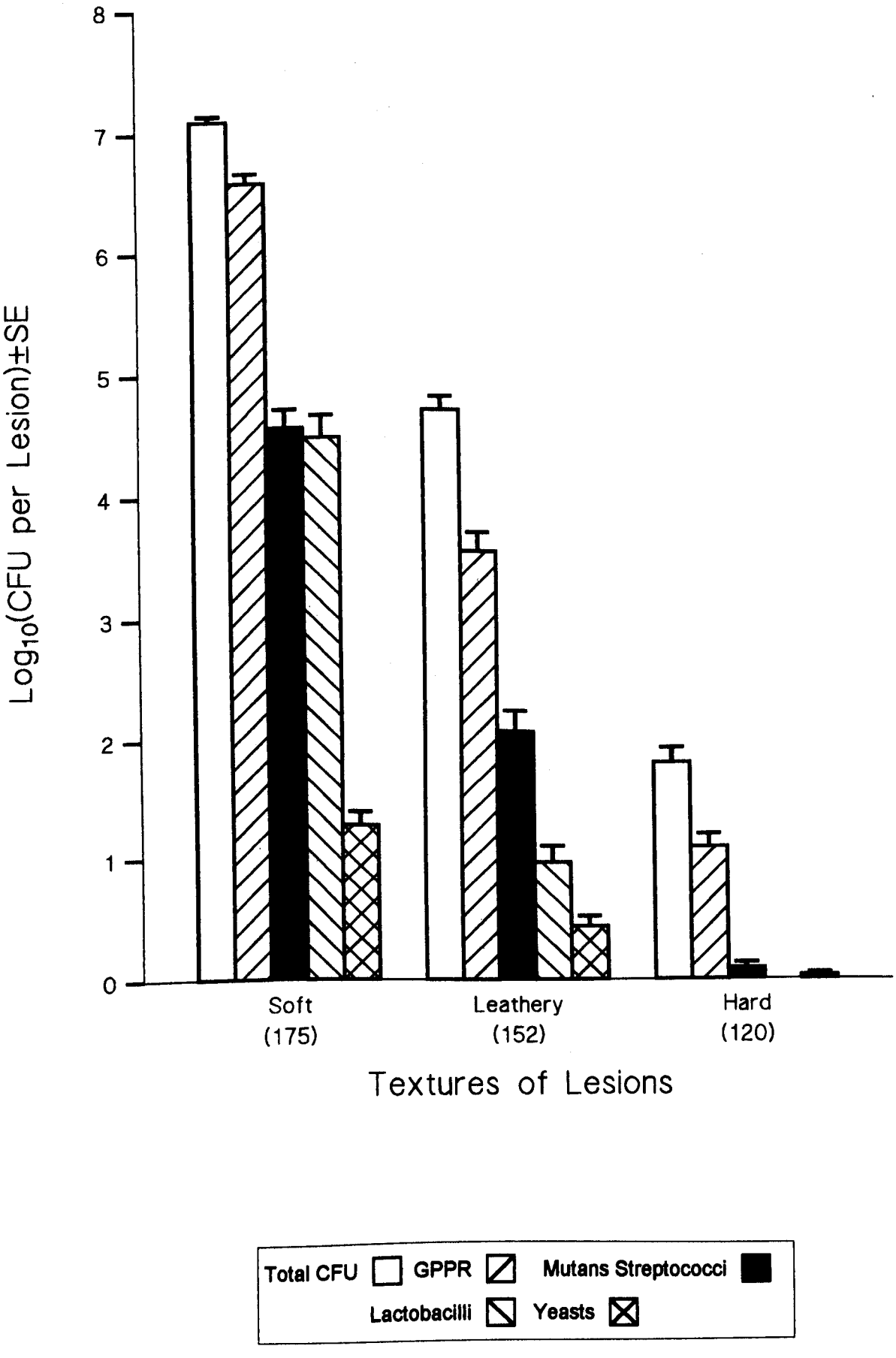
- The Textures of Lesions v The Numbers of Dentine Micro-organisms : V-421
- The Textures of Lesions v The Proportions of Dentine Micro-organisms : V-422
- The Textures of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-423

V-421

The Textures of Lesions

V

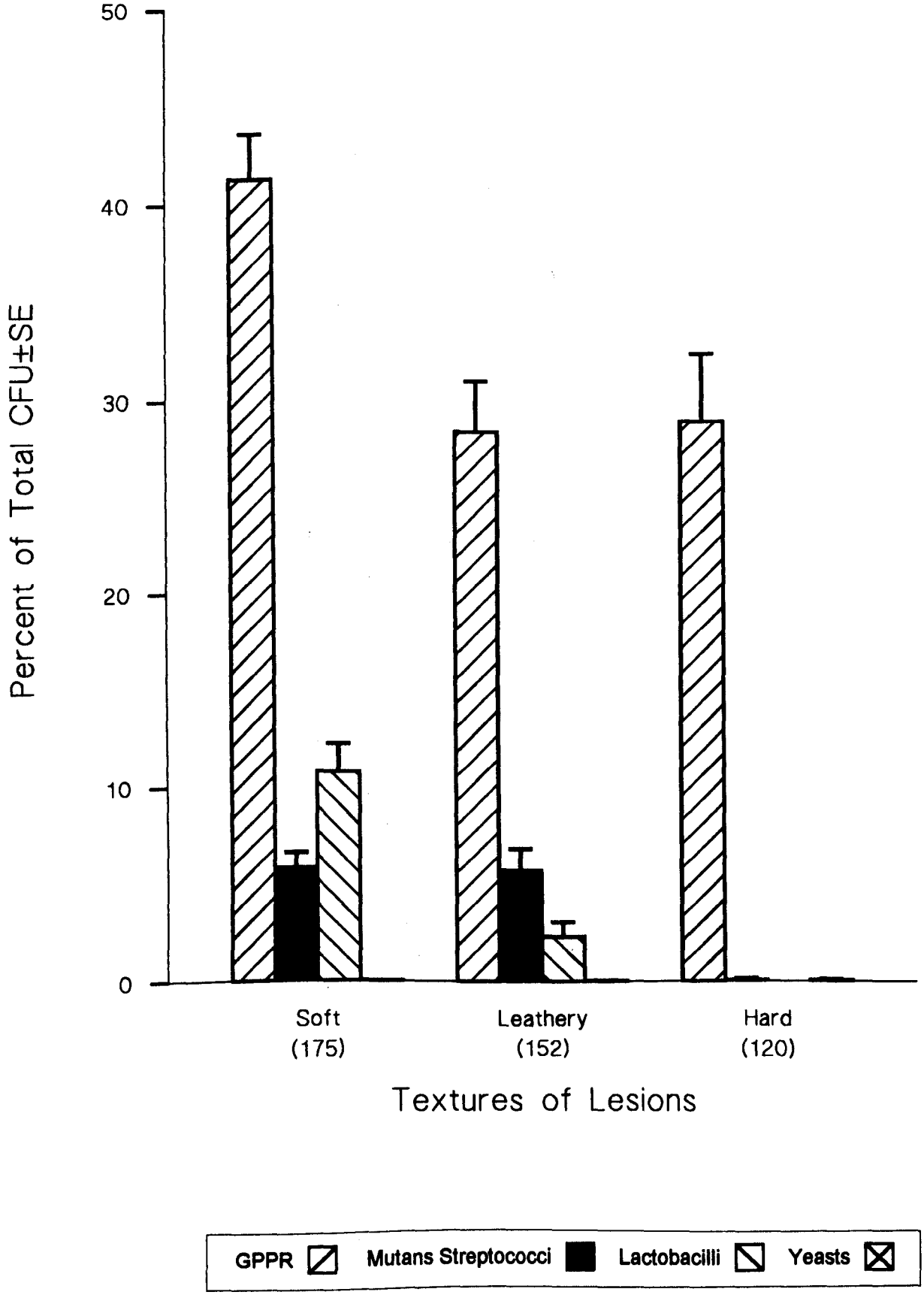
The Numbers of Dentine Micro-organisms



V-421 The Textures of Lesions v The Numbers of Dentine Micro-organisms

The total colony forming units found in the different textured lesions reveal very great differences. Whilst Log_{10} 7.1 ± 0.05 (mean ± standard error) were isolated from the Soft lesions only Log_{10} 4.7 ± 0.11 were found in Leathery lesions and Log_{10} 1.8 ± 0.12 in Hard Lesions ($P < 0.0001$). Again Gram-positive pleomorphic rods were present in the highest numbers in all types of lesion; in the Soft lesions at Log_{10} 6.6 ± 0.08 with Mutans streptococci at Log_{10} 4.6 ± 0.14 and Lactobacilli at Log_{10} 4.5 ± 0.18. The difference in numbers of Lactobacilli in Soft lesions compared with the lower numbers in Leathery lesions (Log_{10} 1.0 ± 0.14) and Hard lesions of zero is also statistically significant ($P < 0.001$). Yeasts were found in fewer numbers (Soft: Log_{10} 1.29 ± 0.11; Leathery : Log_{10} 0.45 ± 0.08 and Hard : Log_{10} 0.03 ± 0.02).

V-422
The Textures of Lesions
V
The Proportions of Dentine Micro-organisms



V-422 The Textures of Lesions v The Proportions of Dentine Micro-organisms

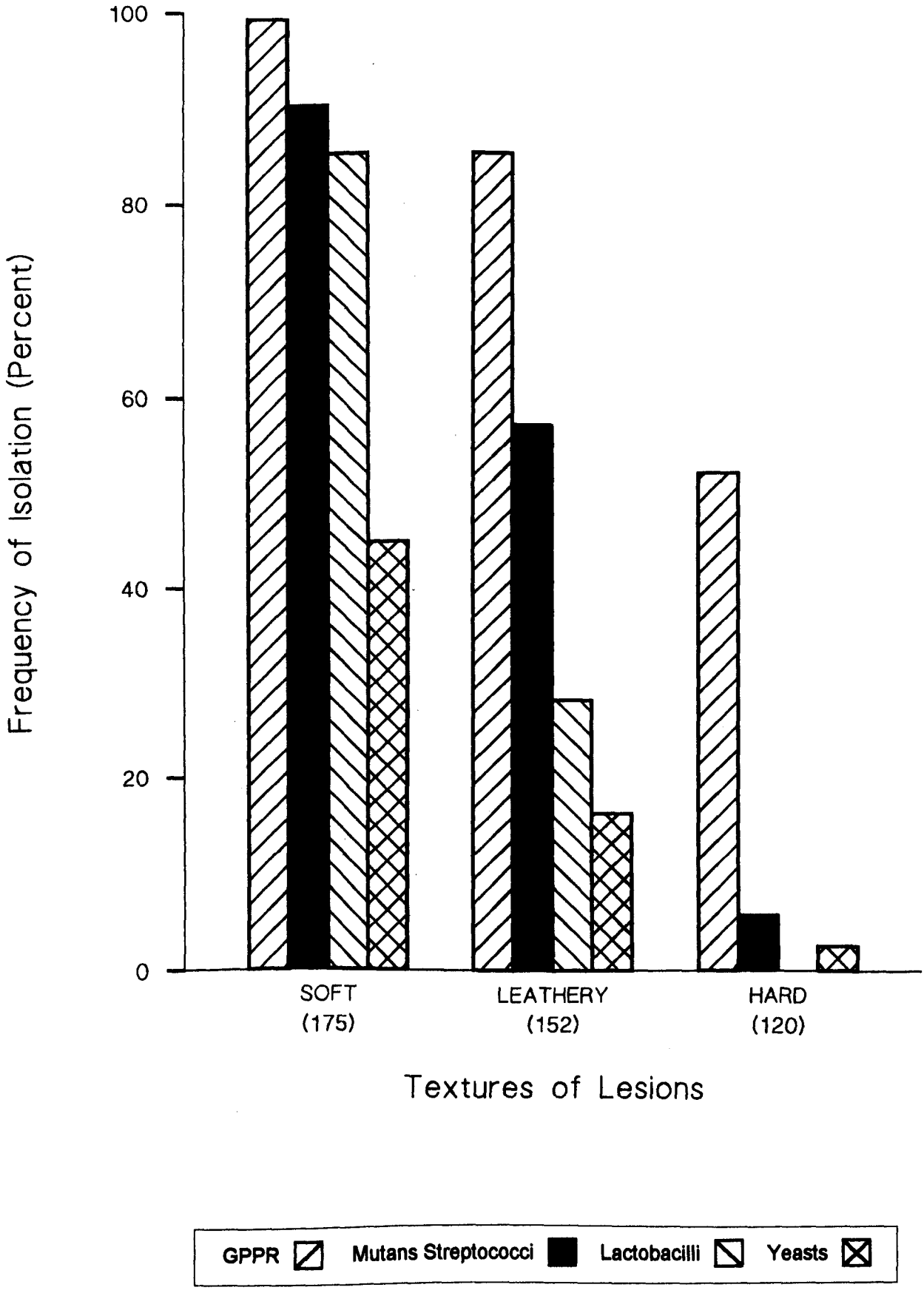
Again, the dominant micro-organisms in all lesions were the Gram-positive pleomorphic rods (Soft: 41 percent; Leathery: 28 percent: and Hard: 29 percent of organisms). Indeed, of the contributions these four types of organisms made to the total colony forming unit counts only Gram-positive pleomorphic rods were present in large numbers in Hard lesions. This histogram also shows the Lactobacilli accounting for 11 percent of organisms from Soft lesions; only 2 percent from Leathery lesions but none from Hard lesions ($P < 0.05$). Mutans streptococci were also virtually absent in Hard lesions (0.1 percent) but form 6 percent of the organisms cultured from both Soft and Leathery dentine ($P < 0.05$).

V-423

The Textures of Lesions

V

The Frequency of Isolation of Dentine Micro-organisms



V-423 The Textures of Lesions v The Frequency of Isolation of Dentine Micro-organisms

99.4 percent of all the Soft lesions contained Gram-positive pleomorphic rods, 85 percent of Leathery lesions but only 52 percent of Hard Lesions. Of Soft lesions, 90 percent also contained Mutans streptococci; 85 percent Lactobacilli and 45 percent Yeasts. Smaller proportions of Leathery lesions were found to be colonised by these three organisms whilst Hard lesions contained even smaller proportions ($P < 0.01$).

V-430 The Locations of Lesions and Their Microbiology

In the three histograms in this section the carious lesions are grouped into the five Locations.

- 122 Lesions were diagnosed to be at the gingival margin;
- 80 were 1 mm or less above it;
- 123 were between >1 and 2 mm;
- 76 were between >2 and 3 mm; and
- 46 were more than 3 mm from the gingival margin

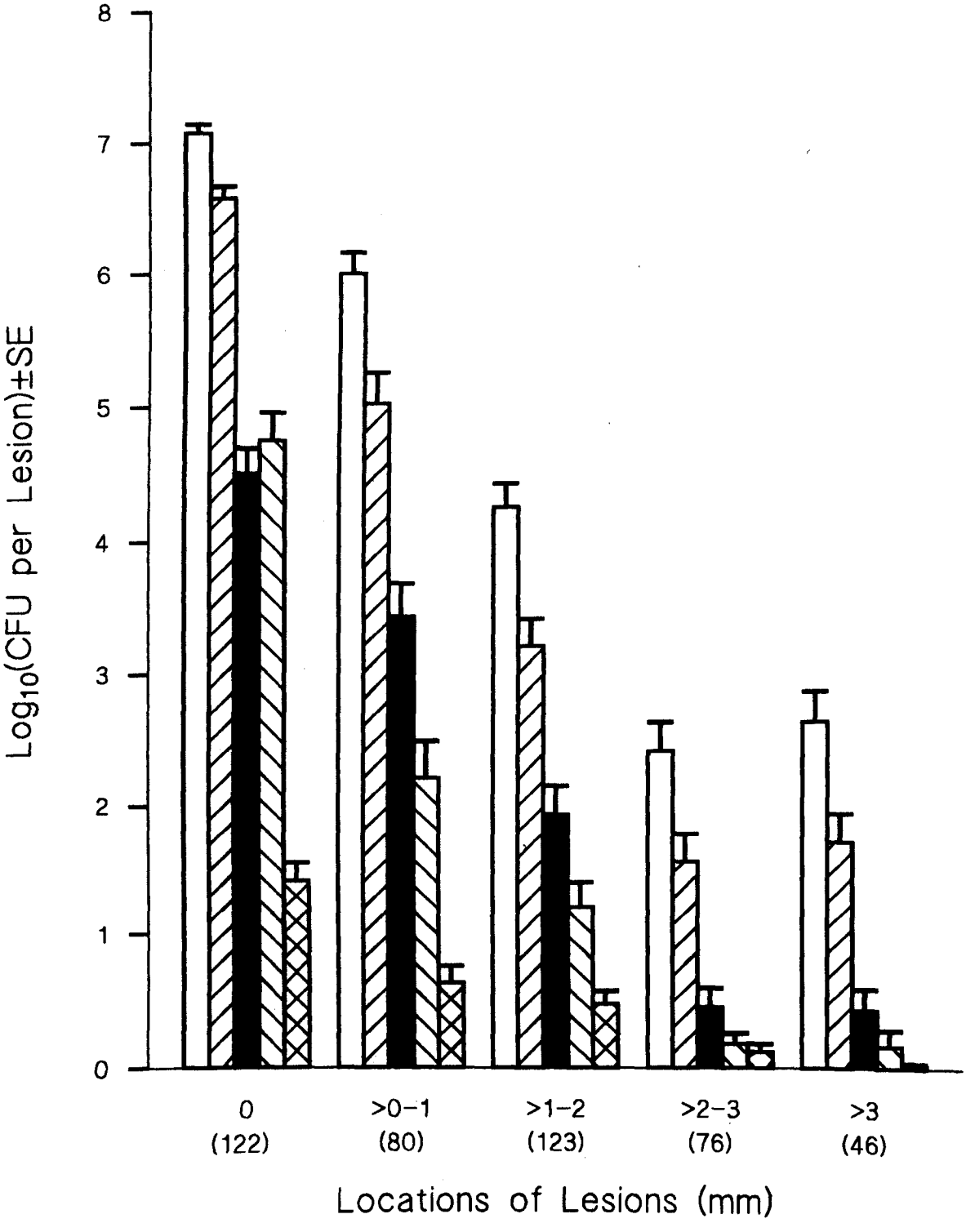
Comparable data are presented as in V-410 and V-420, ie

- The Locations of Lesions v The Numbers of Dentine Micro-organisms : V-431

- The Locations of Lesions v The Proportions of Dentine Micro-organisms : V-432

- The Locations of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-433

V-431
The Locations of Lesions
V
The Numbers of Dentine Micro-organisms



V-431 The Locations of Lesions v The Numbers of Dentine Micro-organisms

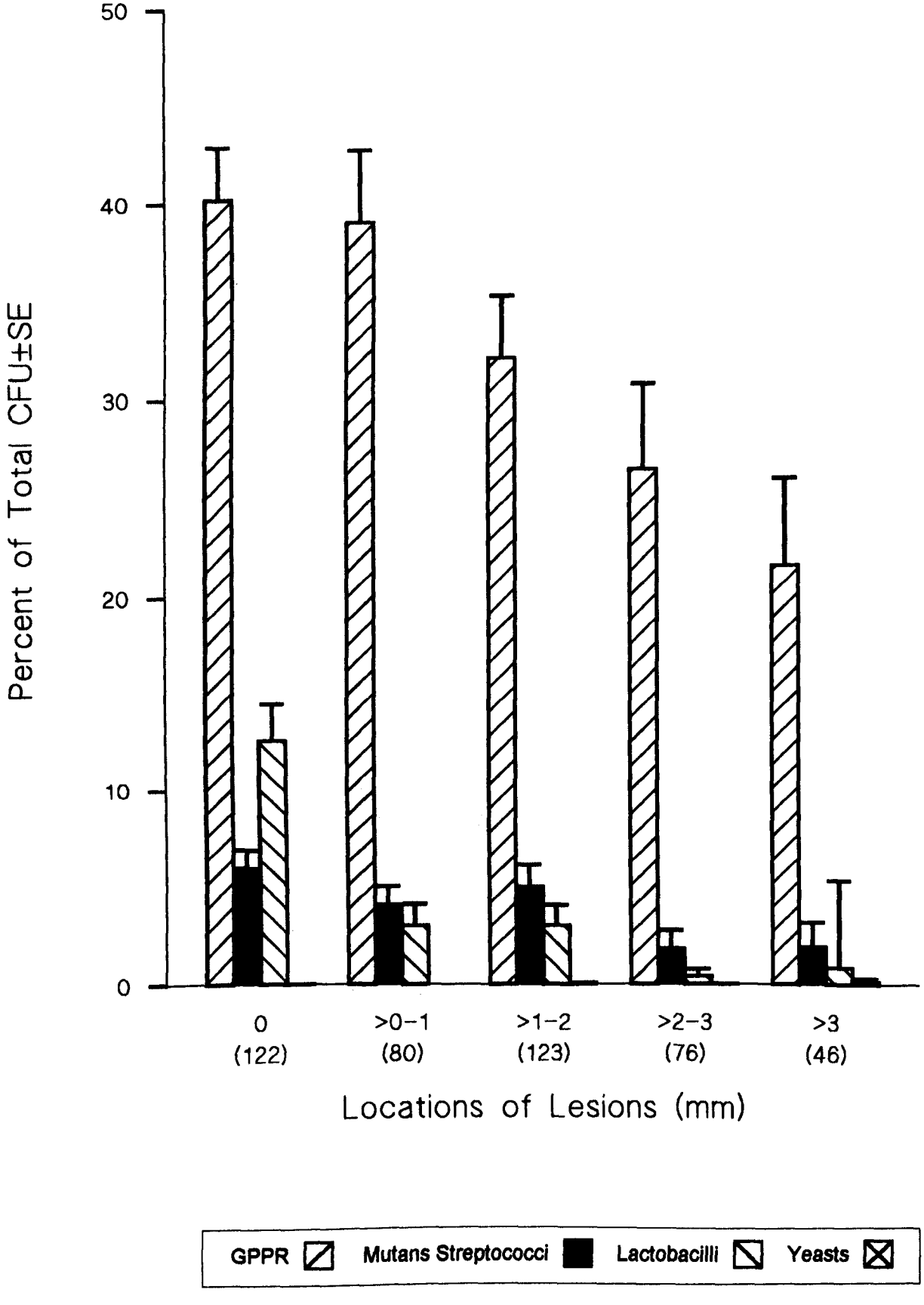
It would seem that the total colony forming units and the total numbers of each of the four types of micro-organisms studied were dramatically reduced in the dentine samples taken from lesions progressively further from the gingival margin. The high numbers of Lactobacilli in lesions located at the gingival margin is such that they exceed the numbers of Mutans streptococci (Log_{10} 4.8 ± 0.2 and Log_{10} 4.5 ± 0.18 respectively), whilst they are found in fewer numbers in all the lesions located further from the gingivae ($P < 0.01$).

V-432

The Locations of Lesions

V

The Proportions of Dentine Micro-organisms



V-432 The Locations of Lesions v The Proportions of Micro-organisms

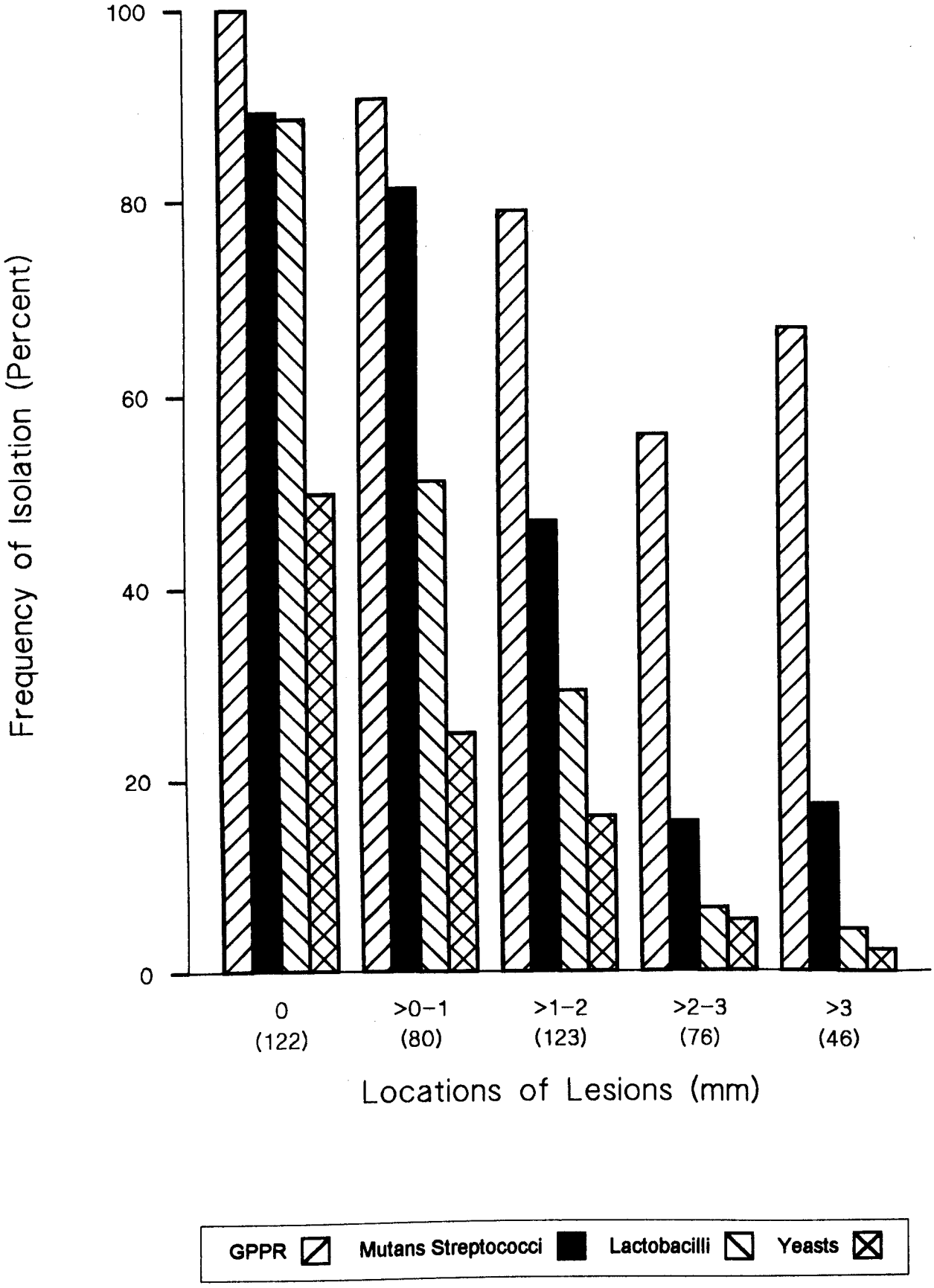
The dominance of Gram-positive pleomorphic rods in all Locations of lesions is clearly revealed in this histogram, for they form from 40 percent in lesions located at the gingival margin reducing to 22 percent in lesions more than 3 mm from it. All other micro-organisms form less than 5 percent of the totals in all groups except for the Lactobacilli and Mutans streptococci which formed 13 percent and 6 percent respectively of the micro-organisms in lesions at the gingival margin ($P < 0.05$).

V-433

The Locations of Lesions

V

The Frequency of Isolation of Dentine Micro-organisms



V-433 The Locations of Lesions v The Frequency of Isolation of Dentine Micro-organisms

The very high Frequency of isolation of all four classes of micro-organisms from lesions located at the gingival margin is clearly revealed in this histogram and the frequencies progressively reduce for each taxon, (except the Gram-positive pleomorphic rods at >3 mm from the gingival margin) as the lesion is judged to be located further and further from the gingival margin. Second only to the Gram-positive pleomorphic rods are the Mutans streptococci which were isolated for 90.3 percent of lesions at the gingiva and 81 percent of those 1 mm or less from it; but dropping to 47 percent in the >1-2 mm band; 6 percent in the >2-3 mm band; and 7 percent in lesions more than 3 mm from the gingival margin. However, whilst only 51 percent of lesions which were less than 1 mm from the margin revealed Lactobacilli, a massive increase to 88.5 percent of those at the gingivae contained them ($P < 0.01$).

V-440 The Cavitations of Lesions and Their Microbiology

The four groupings of the Primary Root Caries lesions in this section are:

- 0.5 mm or less Cavitation (118 lesions);
- >0.5 to 1.5 mm Cavitation (210 lesions);
- >1.5 to 2.5 mm Cavitation (94 lesions); and
- more than 2.5 mm Cavitation (25 lesions).

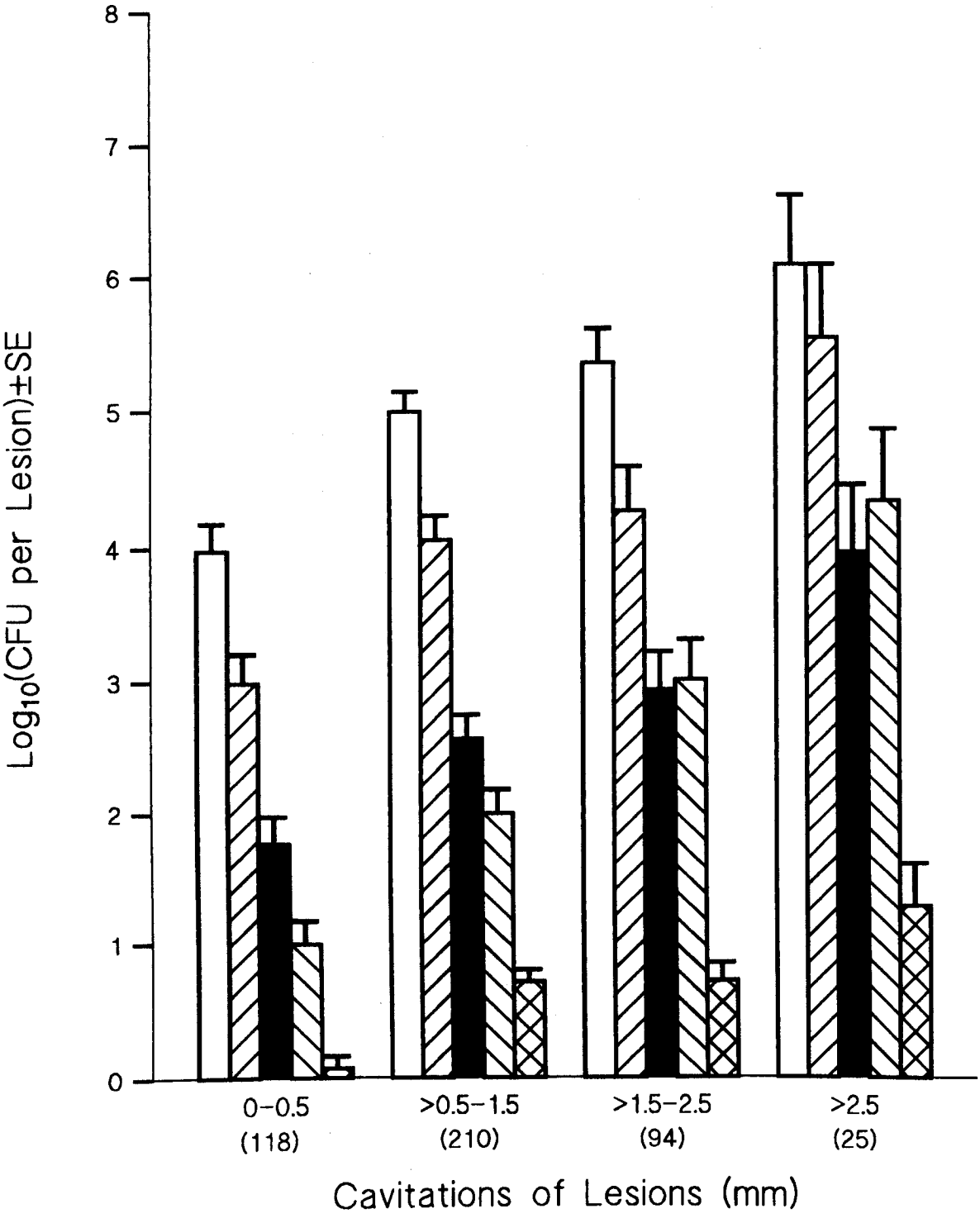
Equivalent to the previous three sections the data are presented as:

- The Cavitations of Lesions v The Numbers of Dentine Micro-organisms : V-441

- The Cavitations of Lesions v The Proportions of Dentine Micro-organisms : V-442

- The Cavitations of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-443

Fig. V-441
The Cavitations of Lesions
V
The Numbers of Dentine Micro-organisms



V-441 The Cavitations of Lesions v The Numbers of Dentine Micro-organisms

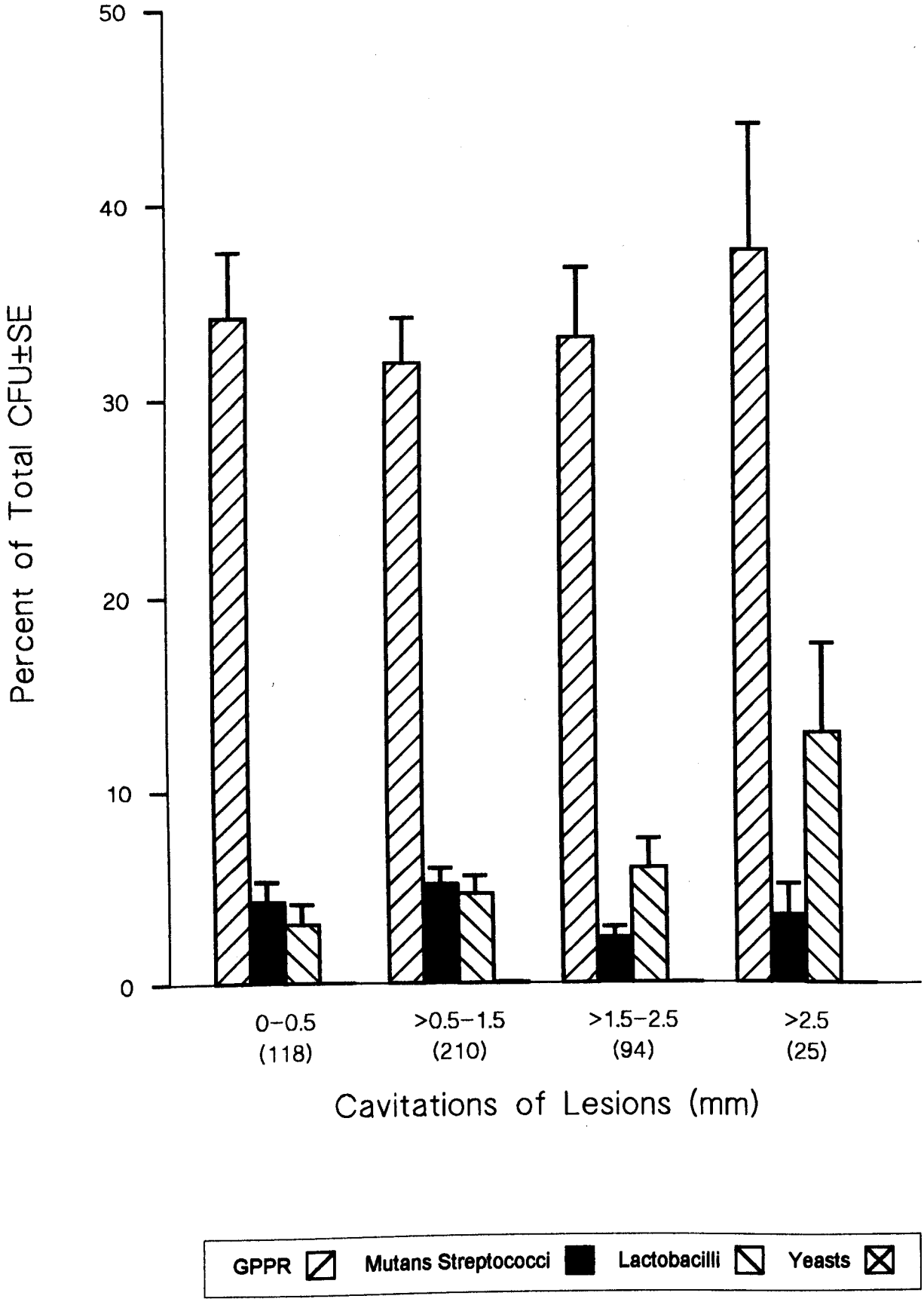
Clearly, the more cavitated the lesions, the higher the total colony forming unit counts are, for they increase from Log_{10} 3.9 ± 0.2 to Log_{10} 6.08 ± 0.53 (mean ± standard error) from the shallowest to the deepest lesions ($P < 0.05$). The dominance of the Gram-positive pleomorphic rods is also evident, so too is the progressively higher numbers of organisms in each category as the lesions develop more cavitated defects. In the 25 lesions which were judged to have lost the greatest depth of surface dentine there are a high number of Lactobacilli (Log_{10} 4.4 ± 0.52) compared to the numbers found in the 118 shallowest lesions (Log_{10} 1.0 ± 0.18) ($P < 0.001$).

V-442

The Cavitations of Lesions

V

The Proportions of Dentine Micro-organisms



V-442 The Cavitations of Lesions v The Proportions of Dentine Micro-organisms

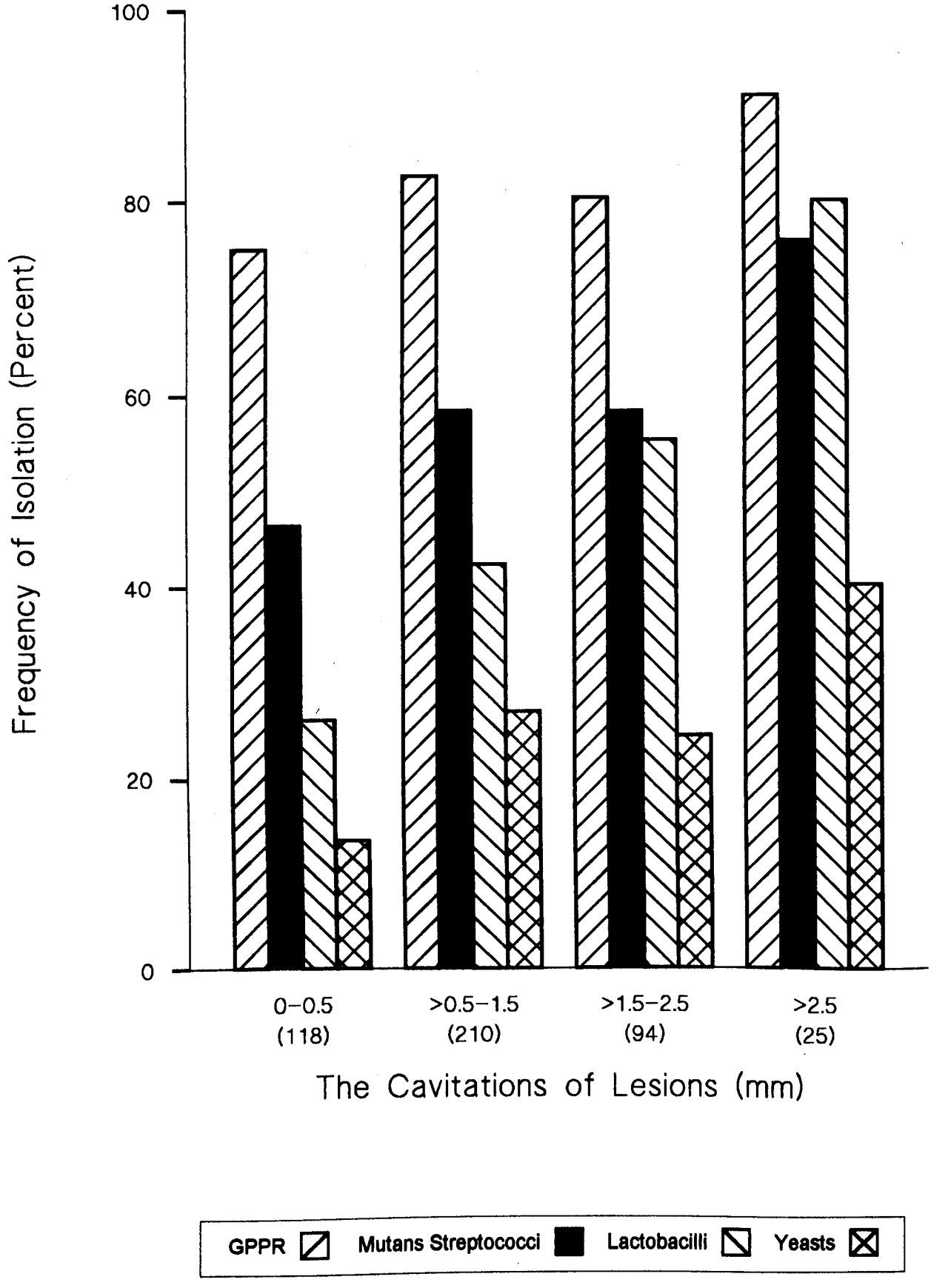
32-37.5 percent of the total colony forming units in all categories of lesions were found to be Gram-positive pleomorphic rods whilst all the other three types accounted for 6 percent or less, with one exception : 13 percent of the micro-organisms from the 25 deepest lesions were found to be Lactobacilli ($P < 0.01$).

V-443

The Cavitations of Lesions

V

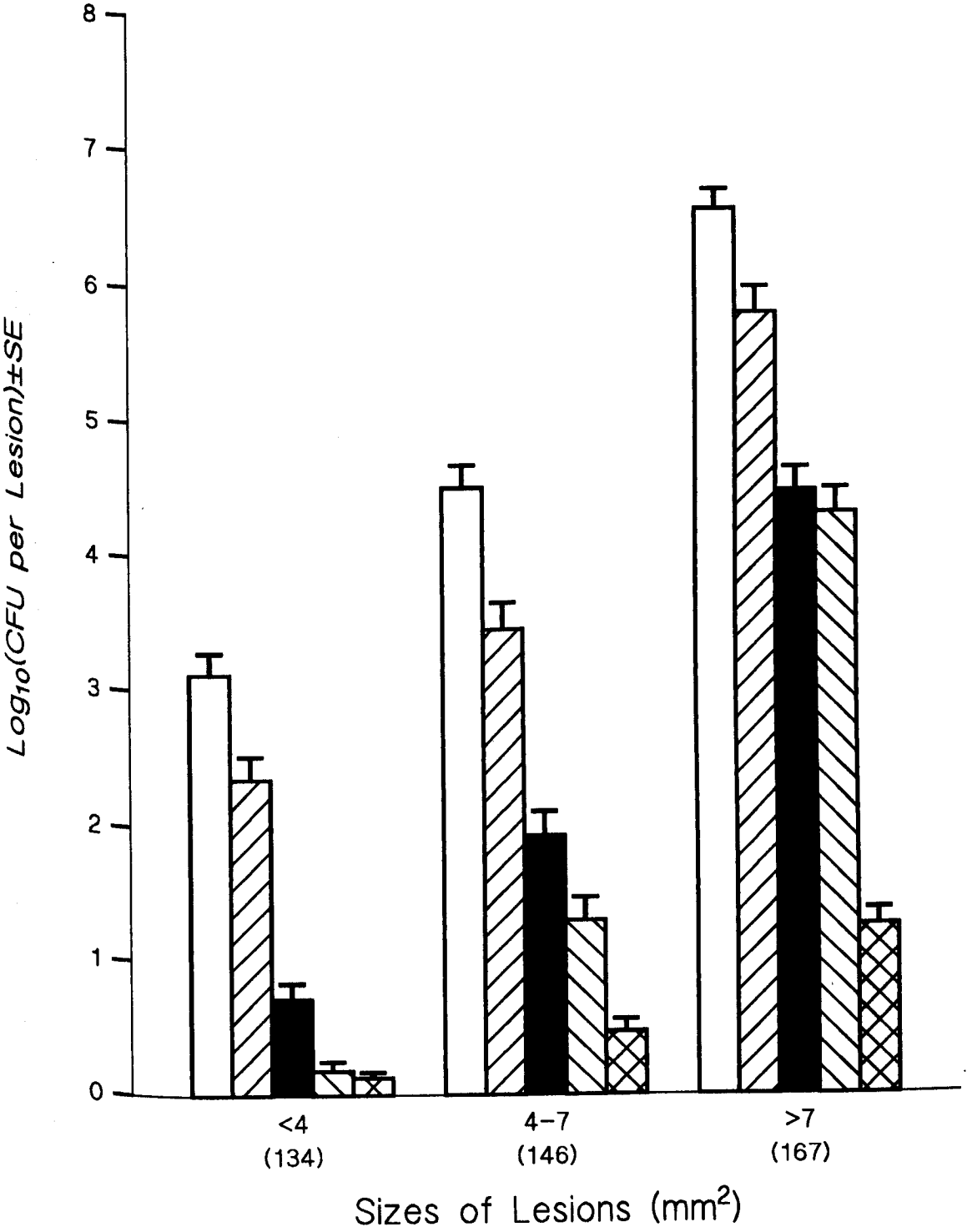
The Frequency of Isolation of Dentine Micro-organisms



V-443 The Cavitations of Lesions v The Frequency of Isolation of Dentine Micro-organisms

The third histogram in this section reveals the high Frequency of isolation of all four taxa of organisms from the deepest lesions (Gram-positive pleomorphic rods: 90 percent, Mutans streptococci : 76 percent; Lactobacilli: 80 percent; and Yeasts: 40 percent) and a constant high frequency from all depths of lesions for Gram-positive pleomorphic rods. The frequency with which Lactobacilli were isolated from the deepest lesions is relatively higher in relation to the other organisms than is evident in the three shallower groups of lesions ($P < 0.05$).

V-451
The Sizes of Lesions
V
The Numbers of Dentine Micro-organisms



V-450 The Sizes of Lesions and Their Microbiology

The Sizes of Lesions have been estimated as the products of their Widths and Heights and do not include any estimate of depth.

- 134 Lesions were diagnosed as being < 4 mm^2 in Size;
- 146 Lesions were diagnosed as being 4 - 7 mm^2 in Size; and
- 167 Lesions were diagnosed as being > 7 mm^2 in Size;

As in the previous sections the results are again presented in three histograms:

- The Sizes of Lesions v The Numbers of Dentine Micro-organisms : V-451
- The Sizes of Lesions v The Proportions of Dentine Micro-organisms : V-452
- The Sizes of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-453

V-451 The Sizes of Lesions v The Numbers of Dentine Micro-organisms

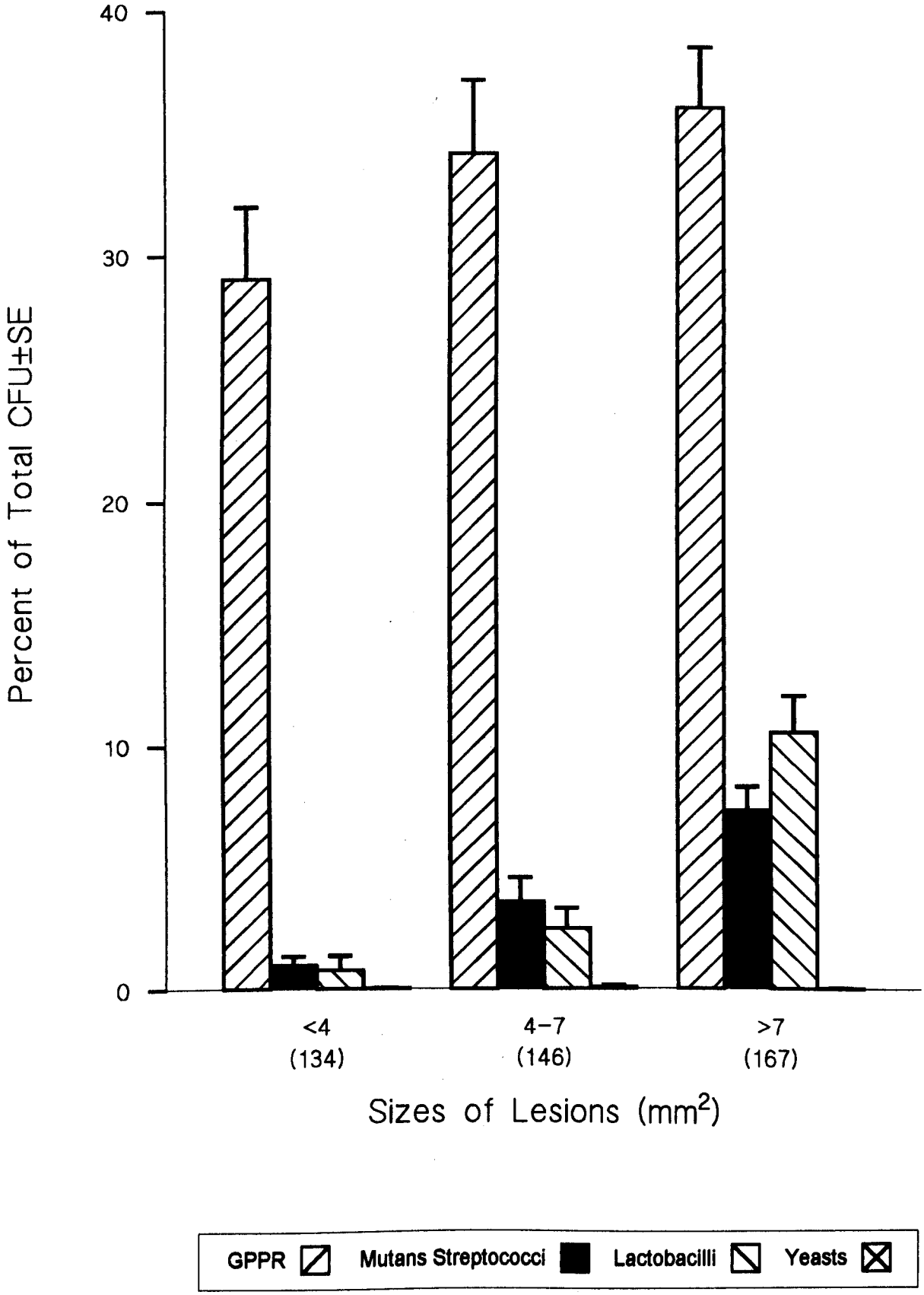
This histogram reveals the dramatic increase in the total colony forming units cultured from the small (Log_{10} 3.1 ± 0.16) to the medium (Log_{10} 4.5 ± 0.17) to the largest (Log_{10} 6.6 ± 0.14) Sizes of Lesions. The same dominance of Gram-positive pleomorphic rods is shown, but far greater numbers of both Mutans streptococci and Lactobacilli were present in the larger lesions than the others ($P < 0.01$).

V-452

The Sizes of Lesions

V

The Proportions of Dentine Micro-organisms



V-452 The Sizes of Lesions v The Proportions of Dentine Micro-organisms

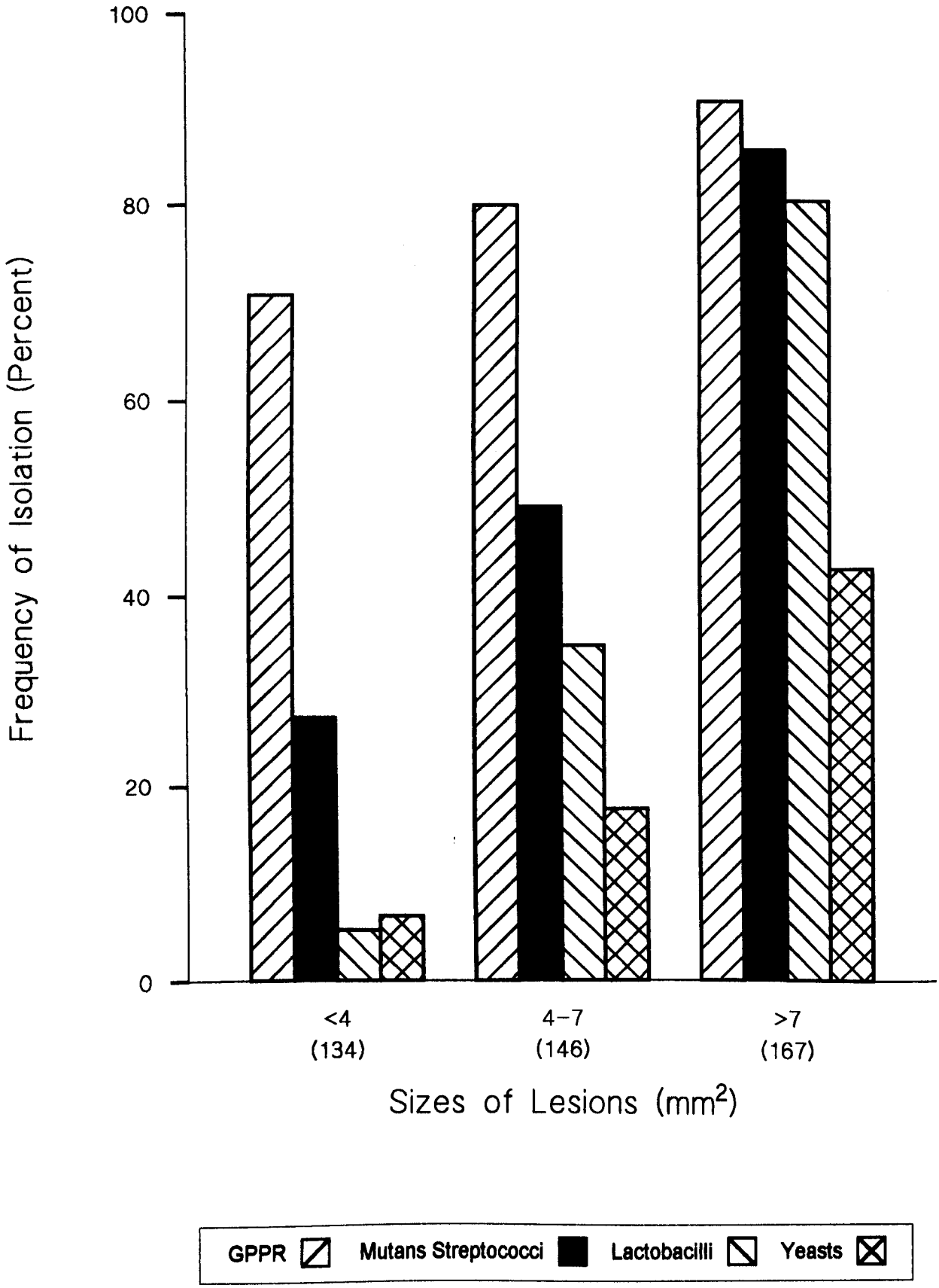
This presentation of the data reveals quite dramatically the predominance of the Gram-positive pleomorphic rods in all Sizes of lesions. Apart from this, the relatively high proportion of Lactobacilli (11 percent) in the largest lesions is clearly evident ($P < 0.01$), whilst the Mutans streptococci formed 7.5 percent in the same group which was also higher than in the other two lesion Sizes ($P < 0.05$).

V-453

The Sizes of Lesions

V

The Frequency of Isolation of Dentine Micro-organisms



V-453 The Sizes of Lesions v The Frequency of Isolation of Dentine Micro-organisms

As well as forming the highest numbers of organisms cultured from all Sizes of lesions Gram-positive pleomorphic rods were also the most frequently isolated, whilst the Mutans streptococci were second most frequently isolated, but they were cultured from lesions with ever increasing frequency, as the Size of the lesions increased, in a more extreme manner than the Gram-positive pleomorphic rods. The other two micro-organisms ; the Lactobacilli and the Yeasts showed a similar increasing frequency as the Sizes increased, though at a lower level ($P < 0.05$).

V-500 The Perceived Treatment Needs of Lesions Related to The Microbiology of Carious Dentine

The numbers and types of micro-organisms isolated from the 447 lesions are related in this section to the Perceived Treatment Needs as defined by the author. Though only four types of treatment have been defined : Restore; Debridement; Chemotherapy; or None, it was decided that it might prove to be helpful to classify those lesions for Restoration into two sections, ie Soft Lesions to be Restored and Leathery Lesions to be Restored.

The five categories involved included:

- Soft Lesions to be Restored (175);
- Leathery Lesions to be Restored (48);
- Lesions to be made Caries Free Only - Debridement (43);
- Lesions to be treated Chemotherapeutically (61) and
- No Treatment (120)

The usual five groupings of micro-organisms are employed. They are presented as:

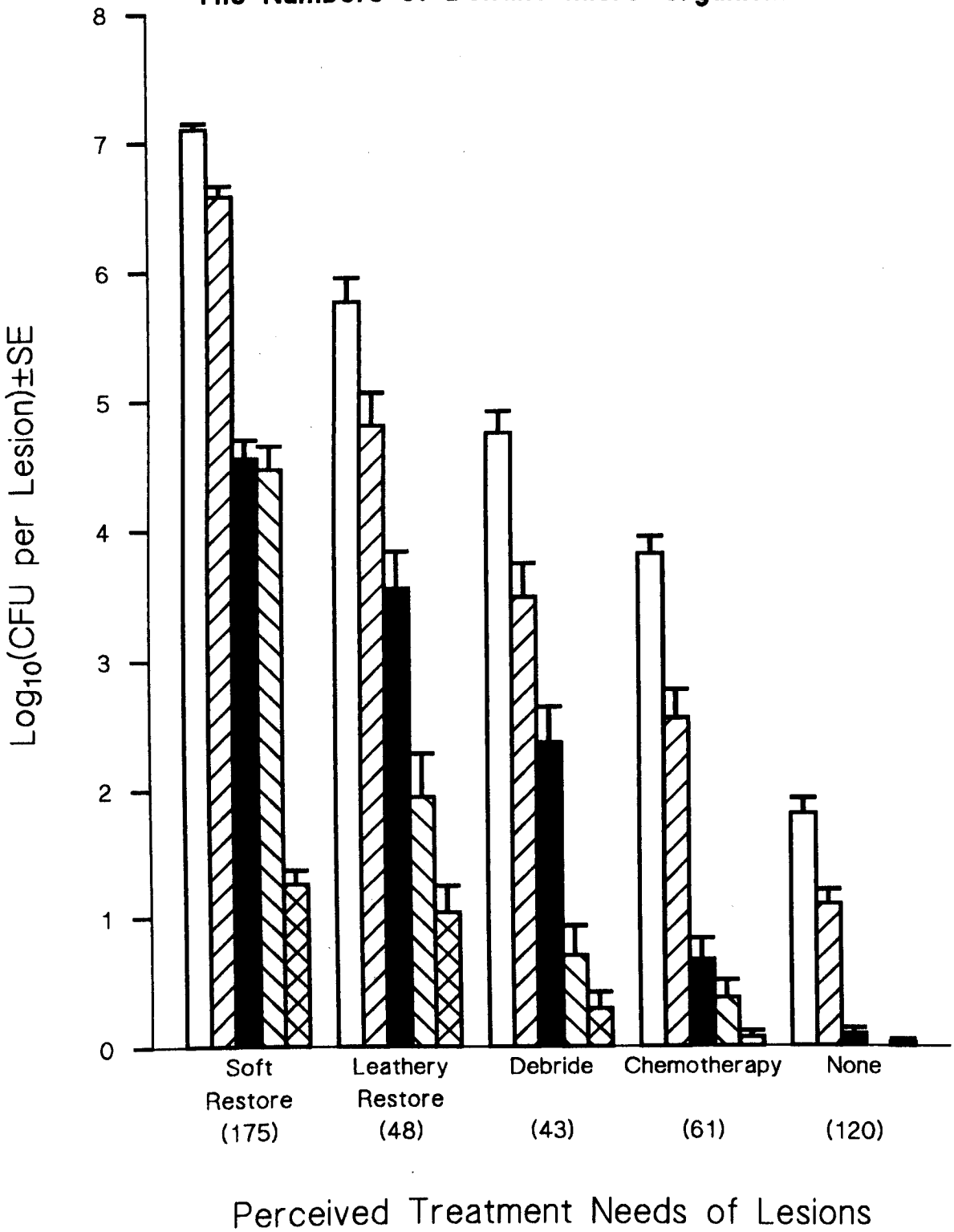
- The Perceived Treatment Needs of Lesions v The Numbers of Dentine Micro-organisms : V-501
- The Perceived Treatment Needs of Lesions v The Proportions of Dentine Micro-organisms : V-502
- The Perceived Treatment Needs of Lesions v The Frequency of Isolation of Dentine Micro-organisms : V-503

V-501

The Perceived Treatment Needs of Lesions

V

The Numbers of Dentine Micro-organisms



V-501 The Perceived Treatment Needs of Lesions v The Numbers of Dentine Micro-organisms

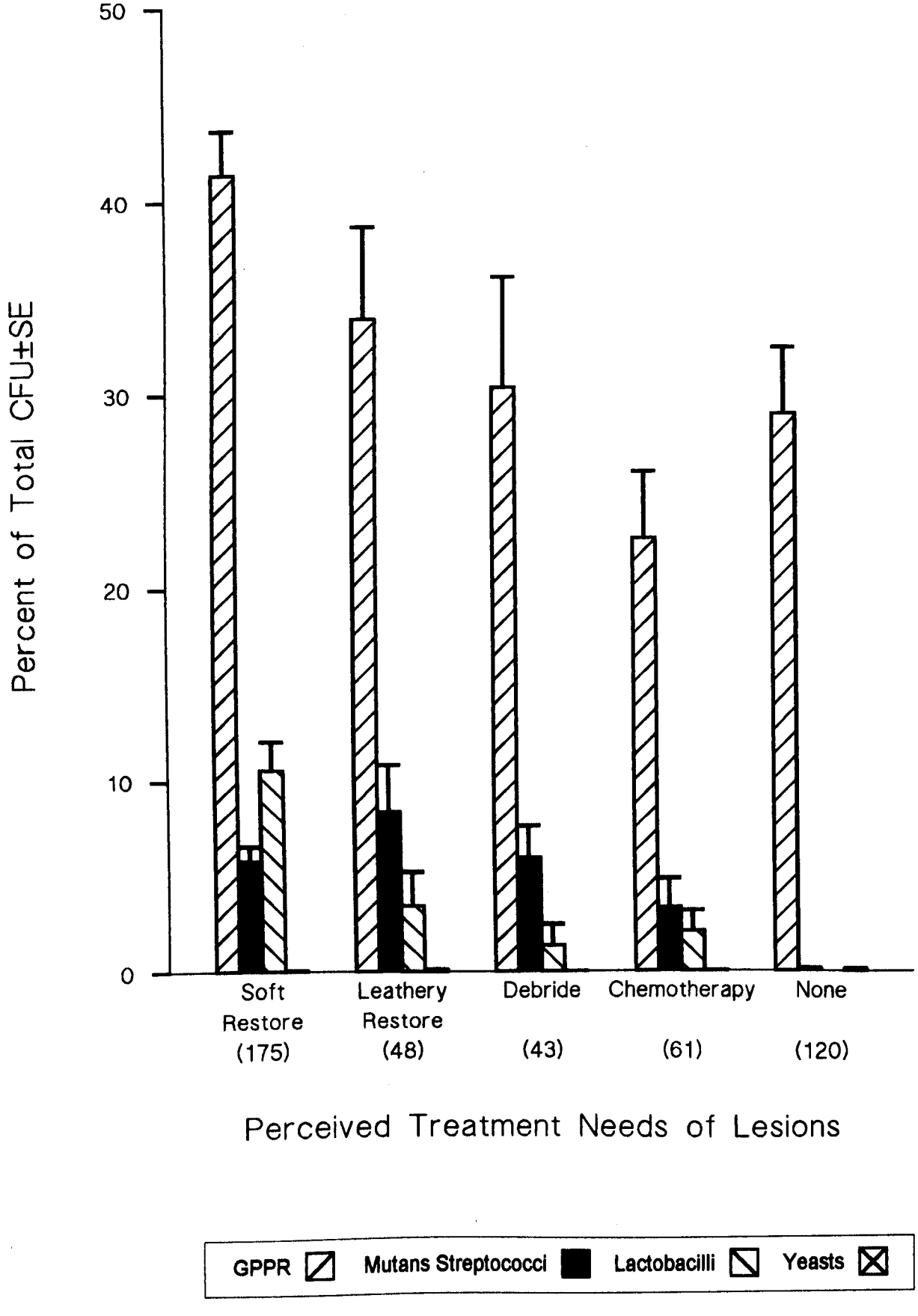
The different treatment strategies chosen for the management of the 447 lesions has resulted in the lesions containing the highest numbers of organisms being subjected to the most radical treatment ($P < 0.01$). The four different types of micro-organisms cultured follow the same pattern, each type being present in fewer numbers in lesions deemed to require less radical treatment ($P < 0.01$).

V-502

The Perceived Treatment Needs of Lesions

V

The Proportions of Dentine Micro-organisms



V-502 The Perceived Treatment Needs of Lesions v The Proportions of Micro-organisms

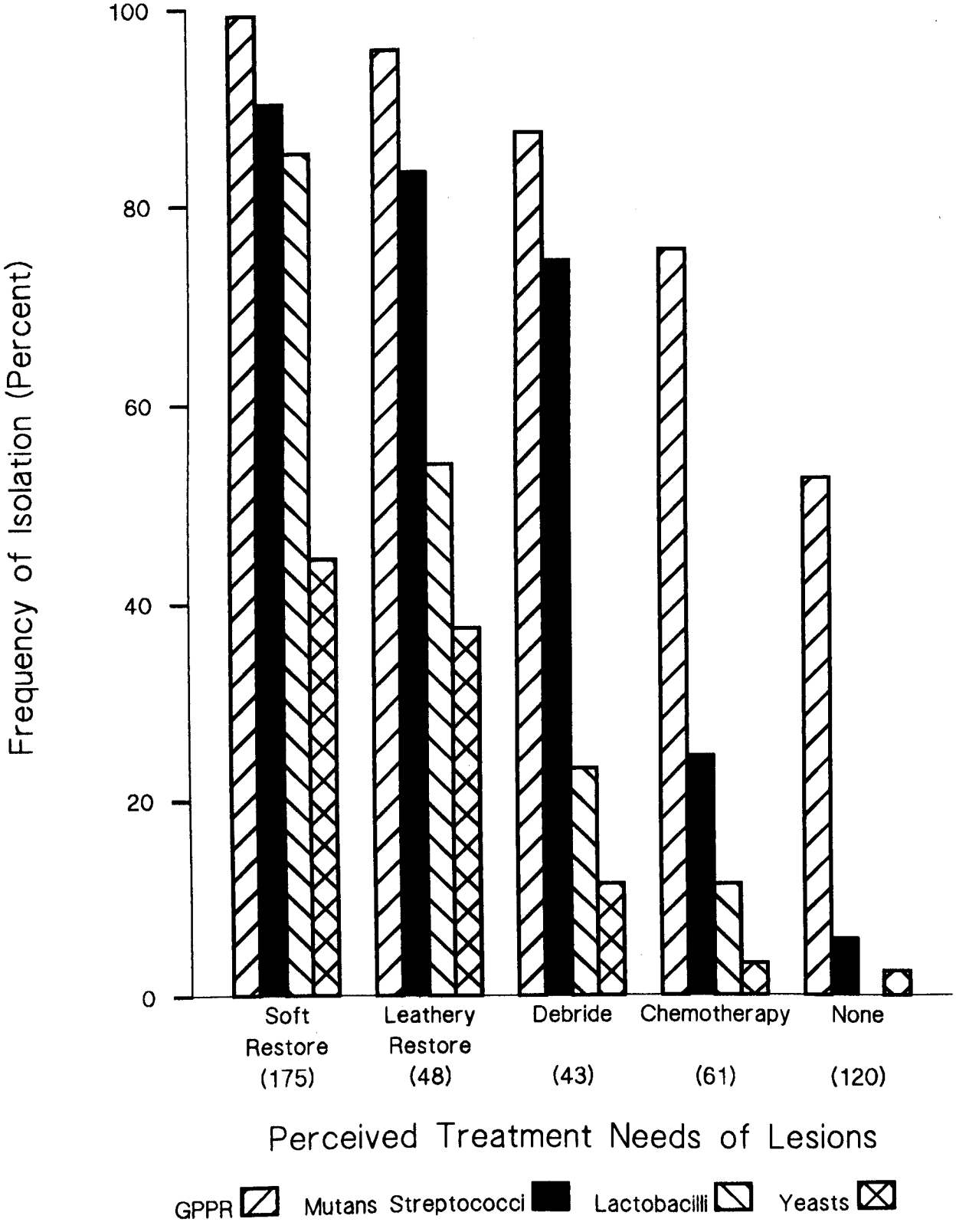
This histogram reveals only that the Soft lesions which were Restored contained higher proportions of Lactobacilli (11 per cent) than the Leathery lesions to be Restored (3.5 percent) ($P < 0.01$). Those lesions for which Chemotherapy was deemed to be required contained fewer Gram-positive pleomorphic rods than those for which no treatment was presented ($P < 0.05$), but the former had higher percentages of Mutans streptococci (3.5 percent) and Lactobacilli (2.2 percent), compared with 0.1 percent and none respectively from those 120 Primary Root Caries lesions deemed to require no treatment ($P < 0.01$).

V-503

The Perceived Treatment Needs of Lesions

V

The Frequency of Isolation of Dentine Micro-organisms



V-503 The Perceived Treatment Needs of Lesions v The Frequency of Isolation of Dentine Micro-organisms

This histogram reveals dramatically the differences between the microbiological characteristics of carious dentine which was considered to require restoration and the other three categories of management. All taxa of micro-organisms were isolated with greater frequency in the Restoration groups than in the others ($P < 0.05$). There was also a significant difference in the characteristics of the microflora of the dentine which it was considered necessary to remove, with or without the restoration of the resultant cavity, compared with the 61 lesions prescribed Chemotherapy and the 120 lesions considered not to need any treatment ($P < 0.05$). The difference is especially obvious in respect of the Mutans streptococci ($P < 0.01$).

V-600 Correlation, Discriminant and Regression Analysis of the Clinical Signs, the Perceived Treatment Needs and the Microbiology of Carious Dentine

The data recorded in V-200 to V-500 show graphically the many inter-relationships between the Clinical Signs, the Perceived Treatment Needs and the Microbiology of Primary Root Caries. In this section the same data are presented to show more precisely the multi-variate analysis and correlation matrix of these variables.

They are presented as:

- The Pooled Within-Groups Correlation Matrix : V-601
- The Combination of Variables that Best Distinguishes Between Different Sub-Groups to Indicate Texture Using Discriminant Analysis : V-602
- The Successful Indication of Texture Group Membership Using 5 Sets of Variables and Discriminant Analysis : V-603
- A Summary of Stepwise Multiple Regression Analysis Results: Indication of the Texture of Lesions : V-604
- The Combination of Variables that Best Distinguishes Between Different Sub-groups to Indicate Perceived Treatment Needs Using Discriminant Analysis : V-605
- The Successful Indication of Perceived Treatment Need Group Membership Using 5 Sets of Variables and Discriminant Analysis : V-606
- A Summary of Stepwise Multiple Regression Analysis Results: Indications of the Perceived Treatment Needs of Lesions : V-607

V-601
Pooled Within-Groups Correlation Matrix

	1 Colour	2 Texture	3 Location	4 Cavitation	5 Size	6 P.T.N	7 Log_{10}(CFU)	8 Log_{10}(GPPR)	9 Log_{10}(MS)	10 Log_{10}(LB)	11 Log_{10}(Y)	12 % GPPR	13 % MS	14 % LB	15 % Y	
1 Colour	1.0000	0.1615**	-0.1696**	0.1362*	0.2515**	-0.1448*	0.2166**	0.2430**	0.1661**	0.2488**	0.2787**	0.0228	-0.0143	0.2887**	-0.0599	Colour
2 Texture	0.1615**	1.0000	-0.7095**	0.2310**	0.5253**	-0.9120**	0.8752**	0.8330**	0.7369**	0.6767**	0.4218**	0.1599**	0.2270**	0.3130**	-0.0489	Texture
3 Location	-0.1696**	-0.7095**	1.0000	-0.2196**	-0.4632**	0.7231**	-0.7130**	-0.6938**	-0.6306**	-0.5992**	-0.3959**	-0.1947**	-0.1410*	-0.2899**	0.0787	Location
4 Cavitation	0.1362*	0.2310**	-0.2196**	1.0000	0.3536**	-0.1532*	0.2321**	0.2322**	0.2273**	0.3451**	0.1793**	0.0049	-0.0538	0.1642**	-0.0100	Cavitation
5 Size	0.2515**	0.5253**	-0.4632**	0.3536**	1.0000	-0.5020**	0.5360**	0.5133**	0.5489**	0.6128**	0.4022**	0.1039	0.1426*	0.4400**	-0.0192	Size
6 P.T.N	-0.1448*	-0.9120**	0.7231**	-0.1532*	-0.5020**	1.0000	-0.8763**	0.8324**	-0.7673**	-0.6355**	-0.4356**	-0.1647**	-0.2742**	-0.2785**	0.0284	P.T.N
7 Log_{10}(CFU)	0.2166**	0.8752**	-0.7130**	0.2321**	0.5360**	-0.8763**	1.0000	0.9434**	0.7871**	0.7236**	0.4869**	0.2259**	0.1890**	0.3018**	-0.0375	Log_{10}(CFU)
8 Log_{10}(GPPR)	0.2430**	0.8330**	-0.6938**	0.2322**	0.5133**	-0.8324**	0.9434**	1.0000	0.7625**	0.7330**	0.4759**	0.4107**	0.1589**	0.3028**	-0.0251	Log_{10}(GPPR)
9 Log_{10}(MS)	0.1661**	0.7369**	-0.6306**	0.2273**	0.5489**	-0.7673**	0.7871**	0.7625**	1.0000	0.6683**	0.4170**	0.1476*	0.4459**	0.2799**	0.0164	Log_{10}(MS)
10 Log_{10}(LB)	0.2488**	0.6767**	-0.5992**	0.3451**	0.6128**	-0.6355**	0.7236**	0.7330**	0.6683**	1.0000	0.5789**	0.2088**	0.1120	0.5793**	-0.0297	Log_{10}(LB)
11 Log_{10}(Y)	0.2787**	0.4218**	-0.3959**	0.1793**	0.4022**	-0.4356**	0.4869**	0.4759**	0.4170**	0.5789**	1.0000	0.0998	0.1501*	0.3696**	0.0973	Log_{10}(Y)
12 % GPPR	0.0228	0.1599**	-0.1947**	0.0049	0.1039	-0.1647**	0.2259**	0.4107**	0.1476*	0.2088**	0.0998	1.0000	-0.0329	0.0502	0.0282	% GPPR
13 % MS	-0.0143	0.2270**	-0.1410*	-0.0538	0.1426*	-0.2742**	0.1890**	0.1589**	0.4459**	0.1120	0.1501*	-0.0329	1.0000	0.0315	0.1060	% MS
14 % LB	0.2887**	0.3130**	-0.2899**	0.1642**	0.4400**	-0.2785**	0.3018**	0.3028**	0.2799**	0.5793**	0.3696**	0.0502	0.0315	1.0000	0.0104	% LB
15 % Y	-0.0599	-0.0489	0.0787	-0.0100	-0.0192	0.0284	-0.0375	-0.0251	-0.0164	-0.0297	0.0973	0.0282	0.1060	0.0104	1.0000	% Y

2-tailed signif: * - .005 ** - .0005

V-601 The Pooled Within-Groups Correlation Matrix

Of these 225 numbers the largest correlation coefficient of 0.9120 is, perhaps not surprisingly, between the Textures of Lesions and their Perceived Treatment Needs (column 2 Line 6). The proportions of Mutans streptococci (Column 13) correlate well with the Perceived Treatment Need (Line 6) at 0.2742; and with Textures (Line 2) at 0.2270, both with $P < 0.005$, whilst Location (Line 3) at 0.1410 and Size (Line 5) at 0.1426 have $P < 0.05$. Location (Column 3), Texture (Column 2) and the Perceived Treatment Needs (Column 6) correlated at $P < 0.05$ with all the groups in this matrix except for the proportions of Yeasts present, but then this latter variable fails to correlate with any of the other groups in the matrix. However, Log_{10} Yeasts (Column 11) correlate with all other groups except the proportions of Gram-positive pleomorphic rods (Line 12) and the proportions of Yeasts (Line 15), whilst the proportions of Gram-positive pleomorphic rods (Column 12) fail to correlate with Colour (Line 1); Cavitation (Line 4); Size (Line 5); Log_{10} Yeasts (Line 11); the proportions of Mutans streptococci (Line 13); the proportions of Lactobacilli (Line 14) and the proportions of Yeasts (Line 15).

V-602

The combination of variables that best distinguishes between different subgroups to indicate texture using discriminant analysis

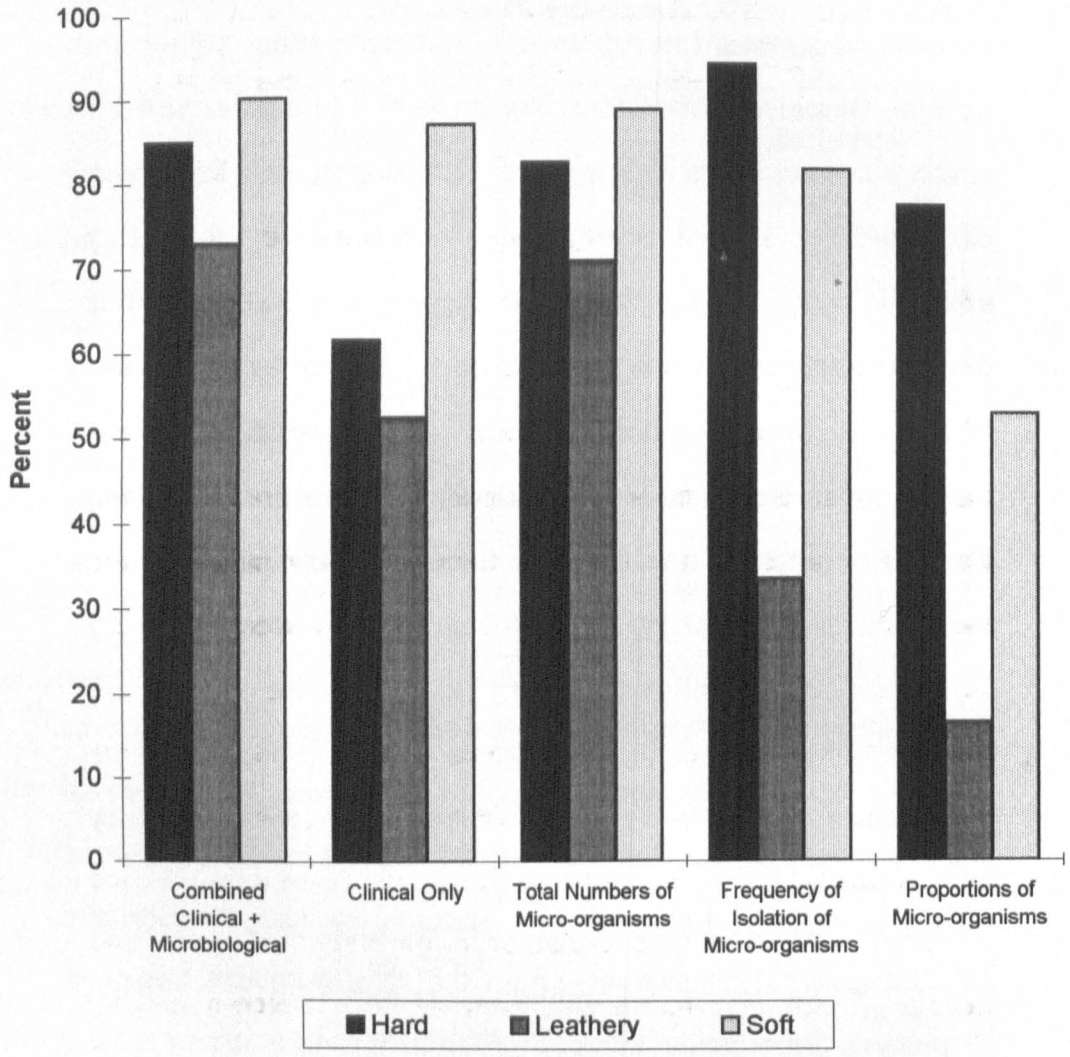
Subgroup	Significant indicator variables ($p<0.001$)	Wilk's Lambda	Non-significant indicator variables
1. Clinical Only			
	Location	0.422	Colour
	Size	0.381	
	Cavitation	0.377	
2. Total Numbers of Micro-organisms (Log_{10})			
	CFU	0.233	Yeasts
	Lactobacilli	0.199	
	GPPR	0.191	
	Mutans Streptococci	0.187	
3. Frequencies of Isolation of Micro-organisms (Freq)			
	Lactobacilli	0.508	
	Mutans Streptococci	0.375	
	GPPR	0.354	
	Yeasts	0.347	
4. Proportions of Micro-organisms (Percent)			
	Lactobacilli	0.891	
	Mutans Streptococci	0.838	
	GPPR	0.807	
	Yeasts	0.801	
5. Combined			
	Log_{10} CFU	0.233	Cavitation
	Location	0.196	Size
	Log_{10} Lactobacilli	0.172	Freq. Lactobacilli
	Log_{10} GPPR	0.166	Freq. Yeasts
	Freq. GPPR	0.154	Percent Yeasts
	Freq. Mutans Streptococci	0.151	
	Log_{10} Mutans Streptococci	0.146	
	Percent Mutans Streptococci	0.142	
	Percent Lactobacilli	0.140	
	Log_{10} Yeasts	0.139	
	Percent GPPR	0.138	
	Colour	0.137	

V-602 The Combination of Variables that Best Distinguishes Between Different Sub-groups to Indicate Texture Using Discriminant Analysis

Since Texture has been shown to correlate most significantly with the Perceived Treatment Needs (V-601), the other variables which correlated to a high level of significance with Texture are perceived to be of importance, not least in respect of epidemiological data. Statistical significance was set at the 0.001 probability level and significant indicator variables were found with respect to Location, Size, and Cavitation (1st Sub-group) whilst Colour was not a significant indicator of the likely Texture of a lesion and of these three, Location was the most important.

The presence of Lactobacilli, followed by Mutans streptococci were the two most important indicators of Texture, notably when the Frequency of isolation and the proportions of the organisms were the subgroups subjected to discriminant analysis (3rd and 4th Sub-groups). However, when the total numbers Log_{10} of Organisms were assessed (Sub-group 2) the numbers of colony forming units and of Lactobacilli were the most significant indicators of the lesion's Texture. The Wilk's Lambda was highest only when the proportions of Micro-organisms were used to classify the Textures of lesions, signifying that the Within-Group variability was large compared with the total variability, ie the proportions of Micro-organisms when used alone were the least reliable indicator variables of Texture.

V-603
Successful Indication of Texture Group Membership Using 5 Sets of Variables and Discriminant Analysis



1. Combined (82.63%)

%	Hard	Leathery	Soft
Hard	**85.0**	15.0	0.0
Leathery	12.1	**73.0**	14.9
Soft	0.0	9.9	**90.1**

2. Clinical Only (68.46%)

%	Hard	Leathery	Soft
Hard	**61.7**	38.3	0.0
Leathery	30.9	**52.6**	16.4
Soft	1.1	12.0	**86.9**

3. Totals Only (80.65%)

%	Hard	Leathery	Soft
Hard	**82.5**	17.5	0.0
Leathery	12.8	**70.9**	16.3
Soft	0.0	11.3	**88.7**

4. Frequency Only (68.49%)

%	Hard	Leathery	Soft
Hard	**94.2**	5.8	0.0
Leathery	41.1	**33.3**	25.5
Soft	0.7	17.6	**81.7**

5. Proportions Only (47.39%)

%	Hard	Leathery	Soft
Hard	**77.5**	1.7	20.8
Leathery	58.9	**16.3**	24.8
Soft	32.4	14.8	**52.8**

Percent of "grouped" cases correctly classified in brackets

V-603 The Successful Indication of Texture Group Membership Using 5 Sets of Variables and Discriminant Analysis

The combinations of variables in each of the five sub-groups given in V-602 which best distinguish Texture were used in this section to indicateto which of the three categories of Texture Primary Root Caries presenting with these characteristics will belong. Using only three clinical criteria (Locations, Size and Cavitation) as shown in the 2nd Group in the histogram and the 2nd Table, 86.9 percent of Soft lesions were correctly indicated, the remaining 13.1 percent were incorrectly indicated as either Leathery (12 percent) or Hard (1.1 percent). With respect to the microbiology of the caries, the Frequency of isolation of Lactobacilli, Mutans streptococci, Gram-positive pleomorphic rods and Yeasts as shown in the 4th Group in the histogram and the 4th Table, provide 94.2 percent correct Indications of all Hard lesions, an accuracy which falls to 77.5 percent if the basis used is the proportions of these micro-organisms. With respect to all Textures the indications were as low as 47.39 percent when only the proportions of the micro-organisms were used as shown in the 5th Table.

V-604
Summary of Stepwise Multiple Regression Analysis Results: Indication of the Texture of Lesions

Dependent Variable	Independent Variables	Beta	R^2
Texture	**1. Clinical only**		
	Location	-0.623 ***	0.580
	Size	0.233 ***	
	2. Total numbers of micro-organisms only		
	Log_{10}(CFU)	0.776 ***	0.770
	Log_{10}(Mutans Streptococci)	0.126 **	
	3. Frequencies of isolation of micro-organisms only (Freq)		
	Mutans Streptococci	0.387 ***	0.621
	Lactobacilli	0.374 ***	
	GPPR's	0.139 ***	
	Yeasts	0.079 *	
	4. Proportions of micro-organisms only (Percent)		
	Lactobacilli	0.298 ***	0.190
	Mutans Streptococci	0.223 ***	
	GPPR's	0.152 **	
	5. Combined 1,2,3 and 4 above		
	Log_{10}(CFU)	0.622 ***	0.800
	Location	-0.136 ***	
	Freq GPPR's	-0.161 ***	
	Log_{10}(GPPR's)	0.257 **	
	Percent Mutans Streptococci	0.068 **	

* $p<0.05$; ** $p<0.01$; *** $p<0.001$

V-604 A Summary of Stepwise Multiple Regression Analysis Results: Indication of the Texture of Lesions

Whilst taking into account any possible correlations between the Locations and the Sizes of lesions, these were the only two clinical signs of Primary Root Caries which significantly correlated by multiple regression analysis with the Texture of the lesions ($P < 0.001$). The Frequency of isolation of Mutans streptococci, Lactobacilli, Gram-positive pleomorphic rods and Yeasts collectively significantly correlated with respect to the Texture and, whilst the Frequency of isolation of Yeasts was significant when the significance level was set at $P < 0.05$, the other three organisms were significant when the significance level was set at $P < 0.001$ and had larger standardised regression coefficients (Betas) than the Yeasts in this third Sub-group.

V-605

The combination of variables that best distinguishes between different subgroups to indicate perceived treatment need using discriminant analysis

Subgroups	Significant indicator variables ($p<0.001$)	Wilk's Lambda	Non-significant indicator variables
1. Clinical Only			
	Texture	0.131	Colour
	Location	0.097	
	Cavitation	0.094	
	Size	0.092	
2. Total Numbers of Micro-organisms (Log_{10})			
	CFU	0.226	
	GPPR	0.197	
	Mutans Streptococci	0.181	
	Lactobacilli	0.174	
	Yeasts	0.172	
3. Frequencies of Isolation of Micro-organisms (Freq)			
	Mutans Streptococci	0.428	
	Lactobacilli	0.300	
	GPPR	0.282	
	Yeasts	0.271	
4. Proportions of Micro-organisms (Percent)			
	Lactobacilli	0.908	Yeasts
	Mutans Streptococci	0.838	
	GPPR	0.802	
5. Combined			
	Texture	0.131	Colour
	Location	0.098	Log_{10} Yeasts
	Freq. Mutans Streptococci	0.082	Percent Lactobacilli
	Log_{10} Mutans Streptococci	0.072	Percent GPPR
	Log_{10} CFU	0.067	Percent Yeasts
	Log_{10} Lactobacilli	0.062	
	Freq. Lactobacilli	0.060	
	Log_{10} GPPR	0.057	
	Freq. GPPR	0.055	
	Cavitation	0.053	
	Percent Mutans Streptococci	0.052	
	Freq. Yeasts	0.051	
	Size	0.051	

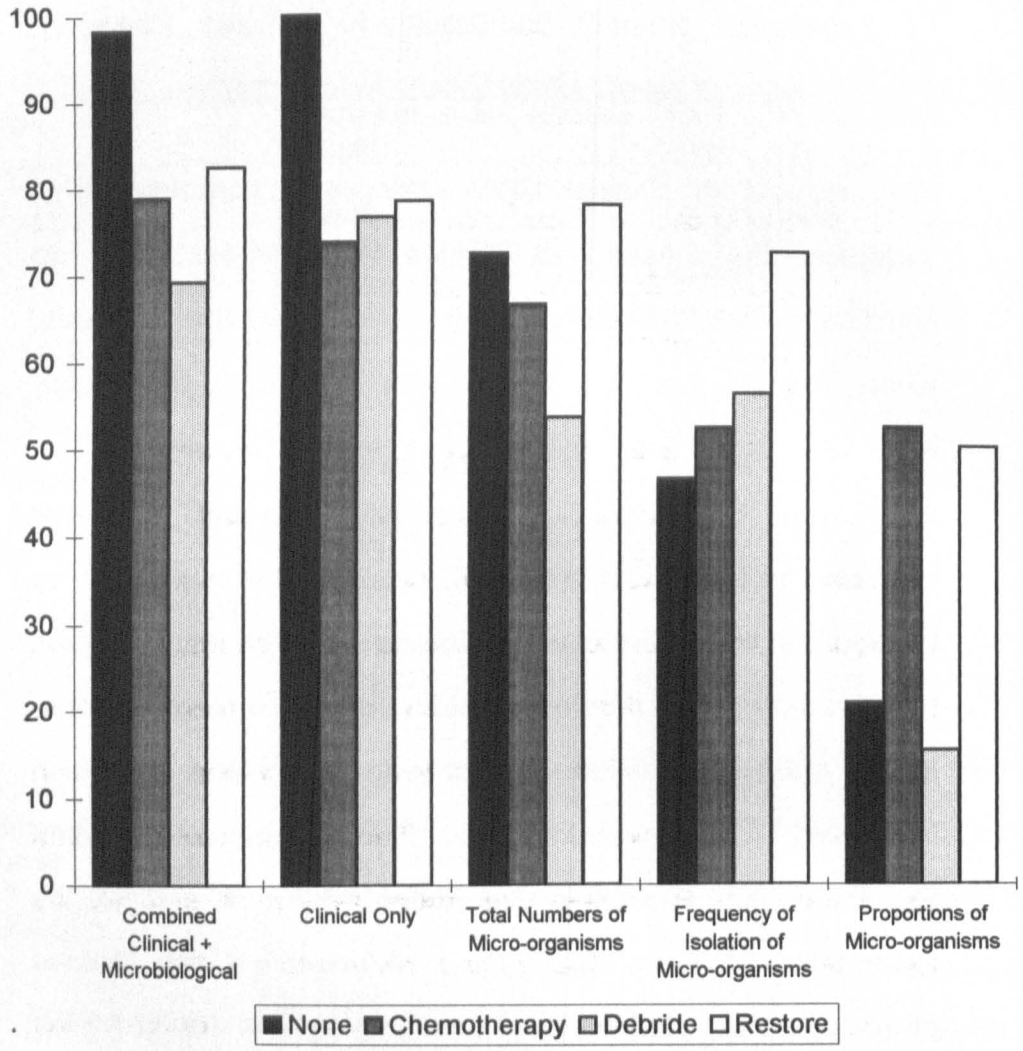
V-605 The Combination of Variables that Best Distinguishes between different sub-groups to Indicate Perceived Treatment Needs Using Discriminant Analysis

With respect to Clinical Signs only as a combination of variables, the Colours of lesions were evidently of no significance as indicators of Perceived Treatment Needs, whilst Texture, Location, Cavitation and Size, in that order, were all significant, the highest being Texture. The significance of Texture was emphasised by the difference between a Wilk's Lambda of 0.131 and the next highest, Location, at 0.097, the others being closer to the latter. This is not surprising since the investigator used these clinical criteria in the subjective clinical assessments of lesions on which Perceived Treatment Needs were based. The sub-groups of the Frequency of isolation of Micro-organisms and of the proportions of Micro-organisms both identified the Mutans streptococci and the Lactobacilli as the two most discriminating variables. The most significant indicator variable in the Sub-group of the total numbers of micro-organisms was Log_{10} of the total numbers of colony forming units.

When all the four sub-groups of data are combined the Texture and the Location of lesions, especially Texture at 0.131 outstripped the other criteria whilst Colour remained in the non-significant category.

V-606

Successful Indication of Perceived Treatment Need Group Membership Using 5 Sets of Variables and Discriminant Analysis



1. Combined (85.36%)

%	N	C	D	R
None	**98.3**	0.0	1.7	0.0
Chemotherapy	0.0	**78.9**	21.1	0.0
Debride	0.0	28.2	**69.2**	2.6
Restore	0.0	2.1	15.5	**82.4**

2. Clinical Only (83.45%)

%	N	C	D	R
None	**100**	0.0	0.0	0.0
Chemotherapy	0.0	**73.8**	26.2	0.0
Debride	0.0	23.3	**76.7**	0.0
Restore	0.0	2.2	19.3	**78.5**

3. Totals Only (72.46%)

%	N	C	D	R
None	**72.5**	23.3	4.2	0.0
Chemotherapy	10.5	**66.7**	19.3	3.5
Debride	5.1	30.8	**53.8**	10.3
Restore	0.0	4.8	17.1	**78.1**

4. Frequency Only (60.55%)

%	N	C	D	R
None	**46.7**	47.5	5.8	0.0
Chemotherapy	22.8	**52.6**	17.5	7.0
Debride	5.1	23.1	**56.4**	15.4
Restore	0.5	5.3	21.4	**72.7**

5. Proportions Only (38.46%)

%	N	C	D	R
None	**20.8**	57.5	1.7	20.0
Chemotherapy	22.8	**52.6**	5.3	19.3
Debride	12.8	43.6	**15.4**	28.2
Restore	15.0	24.6	10.2	**50.3**

Percent of 'grouped' cases correctly classified in brackets

V-606 The Successful Indication of Perceived Treatment Need Group Membership Using 5 Sets of Variables and Discriminant Analysis

As revealed in the 2nd Group in the Histogram and the 2nd Table, the use of Clinical Criteria only produced a high level of indication of Perceived Treatment Needs at 83.45 percent for the four treatment categories. This is not perhaps surprising since the investigator used such clinical criteria in the subjective clinical assessments of lesions on which Perceived Treatment Needs were based. The microbiological data correlated with Perceived Treatment Needs at significantly lower levels but, of these, the total numbers of Micro-organisms proved to be the best combination of variables averaging 72.46 percent; behind this group are the Frequency of isolation of Micro-organisms averaging 60.55 percent, whilst the proportions of the different Micro-organisms are shown as being much less effective in indicating Perceived Treatment Needs at 38.46 percent.

V-607
Summary of Stepwise Multiple Regression Analysis Results: Indication of the Perceived Treatment Needs of Lesions

Dependent Variable	Independent Variables	Beta	R^2
Perceived Treatment Needs	**1. Clinical only**		
	Texture	0.931 ***	0.867
	2. Total numbers of micro-organisms only		
	Log_{10}(CFU)	0.768 ***	0.753
	Log_{10}(Mutans Streptococci)	0.123 **	
	3. Frequencies of isolation of micro-organisms only (Freq)		
	Mutans Streptococci	0.415 ***	0.624
	Lactobacilli	0.330 ***	
	GPPR's	0.160 ***	
	Yeasts	0.082 ***	
	4. Proportions of micro-organisms only (Percent)		
	Lactobacilli	0.267 ***	0.166
	Mutans Streptococci	0.265 **	
	GPPR's	0.137 *	
	5. Combined 1,2,3 and 4 above		
	Texture	0.734 ***	0.888
	Log_{10}(CFU)	0.250 **	
	Percent Mutans Streptococci	0.047 ***	
	Log_{10}(Lactobacilli)	-0.245 ***	
	Freq Lactobacilli	0.173 *	
	Percent Lactobacilli	0.045	

* p<0.05; ** p<0.01; *** p<0.001

V-607 A Summary of Stepwise Multiple Regression Analysis Results: Indications of the Perceived Treatment Needs of Lesions

This table summarises the data presented in the earlier parts of this section. All the factors that have been found to be of high significance are clearly demonstrated and, since these have already been highlighted, further reference to them will not be made here.

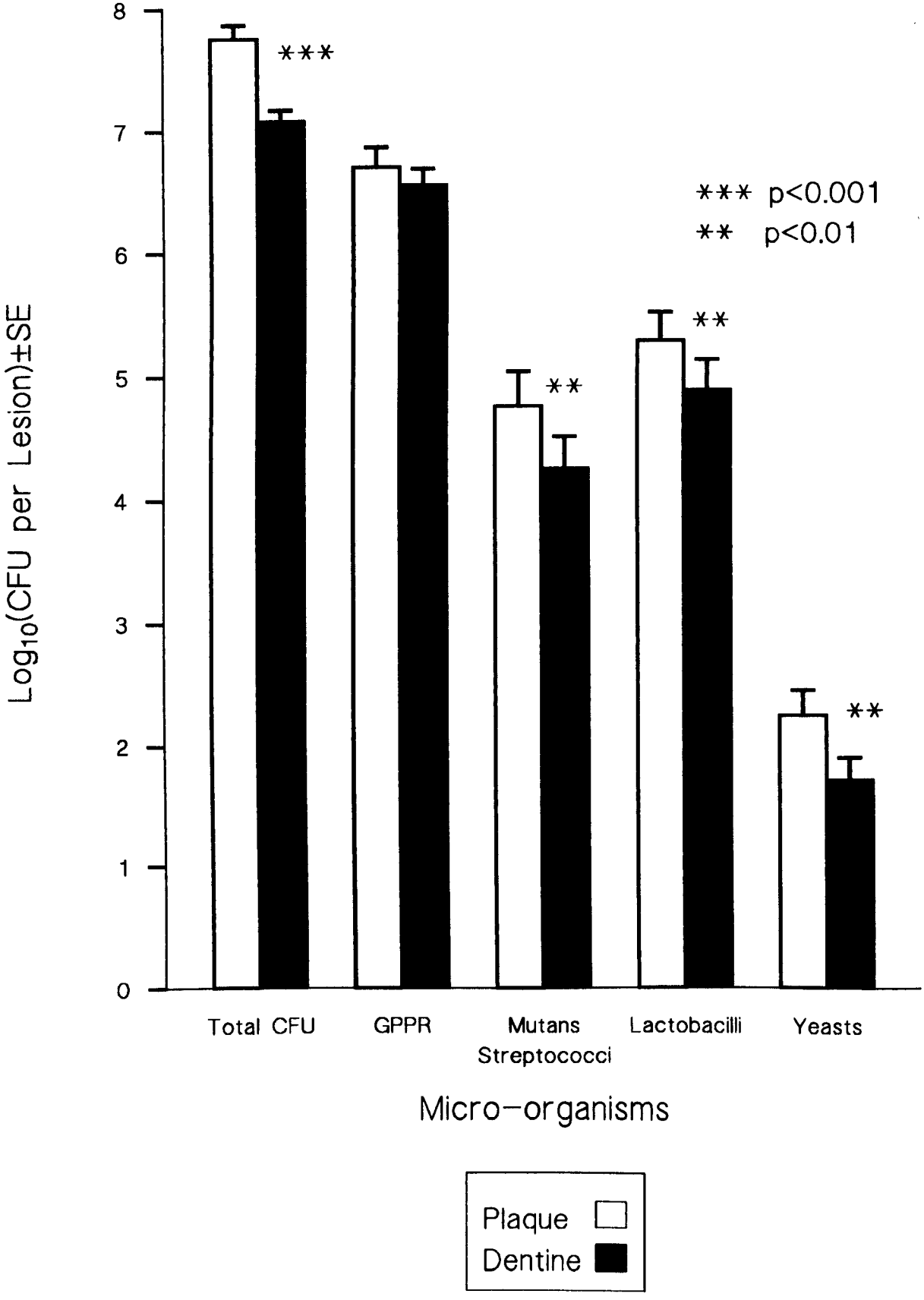
V-700 The Microbiology of Carious Dentine Related to the Microbiology of the Overlying Plaque

The numbers and taxa of micro-organisms isolated from 81 lesions in 52 patients and from the plaque overlying each of these lesions is reported in this section with respect to the same groups of micro-organisms listed in V-400 and in Section IV. They are also presented in the same three ways, ie:-

- The Numbers of Dentine Micro-organisms v The Numbers of Plaque Micro-organisms : V-701
- The Proportions of Dentine Micro-organisms v The Proportions of Plaque Micro-organisms : V-702
- The Frequency of Isolation of Dentine Micro-organisms v The Frequency of Isolation of Plaque Micro-organisms : V-703

V-701

The Numbers of Dentine Micro-organisms V The Numbers of Plaque Micro-organisms

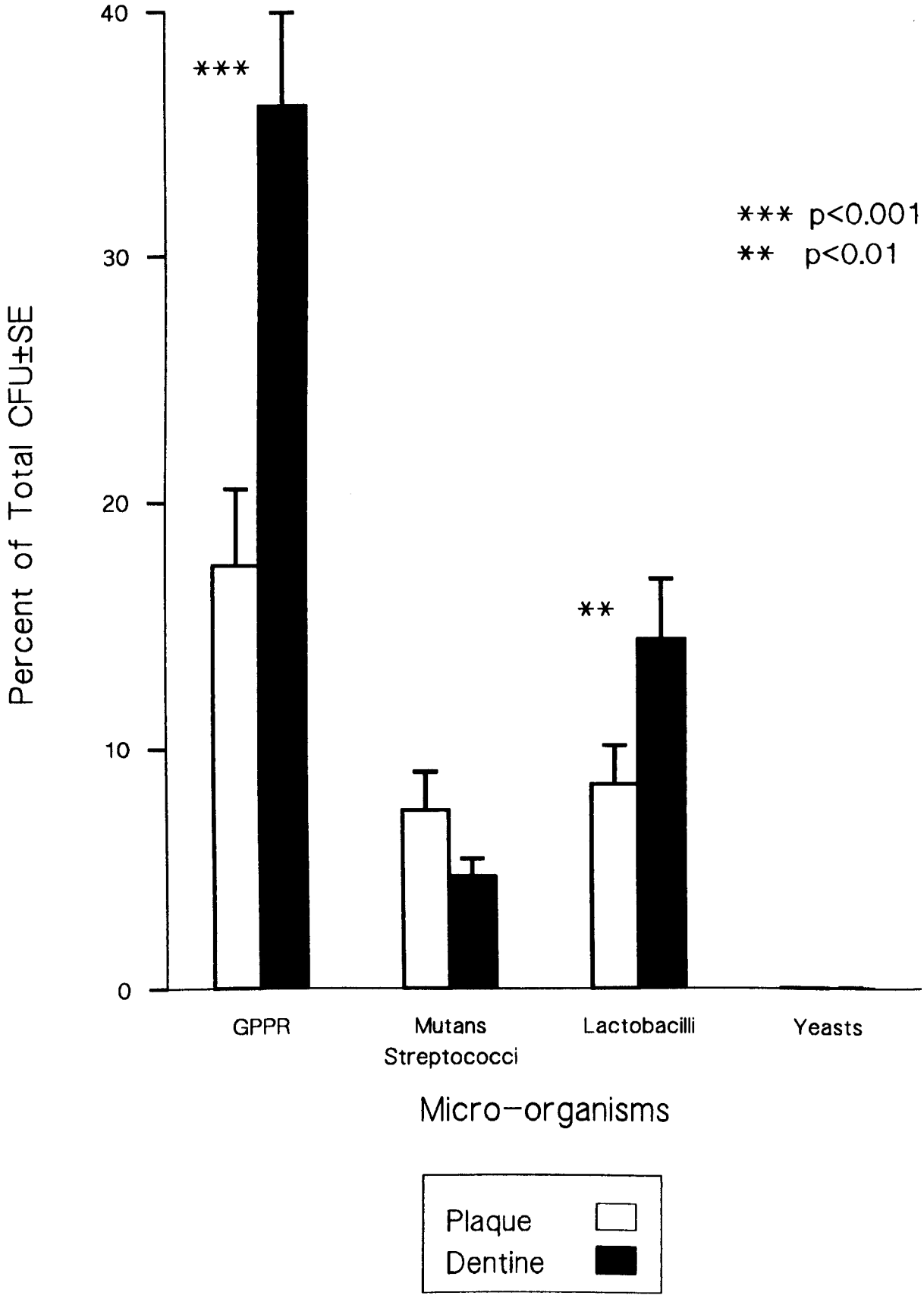


V-701 The Numbers of Dentine Micro-organisms v The Numbers of Plaque Micro-organisms

Though the Log_{10} scale fails to demonstrate graphically the significant variations in the numbers of micro-organisms isolated from carious lesions themselves and from the plaque overlying them, these differences are clearly evident from the figures relating to the total colony forming units ($P < 0.001$); Mutans streptococci ($P < 0.01$); Lactobacilli ($P < 0.01$) and Yeasts ($P < 0.01$). Only the numbers of Gram-positive pleomorphic rods cultured from dentine and plaque respectively failed to reveal highly significant differences.

V-702

The Proportions of Dentine Micro-organisms V The Proportions of Plaque Micro-organisms



V-702 The Proportions of Dentine Micro-organisms v The Proportions of Plaque Micro-organisms

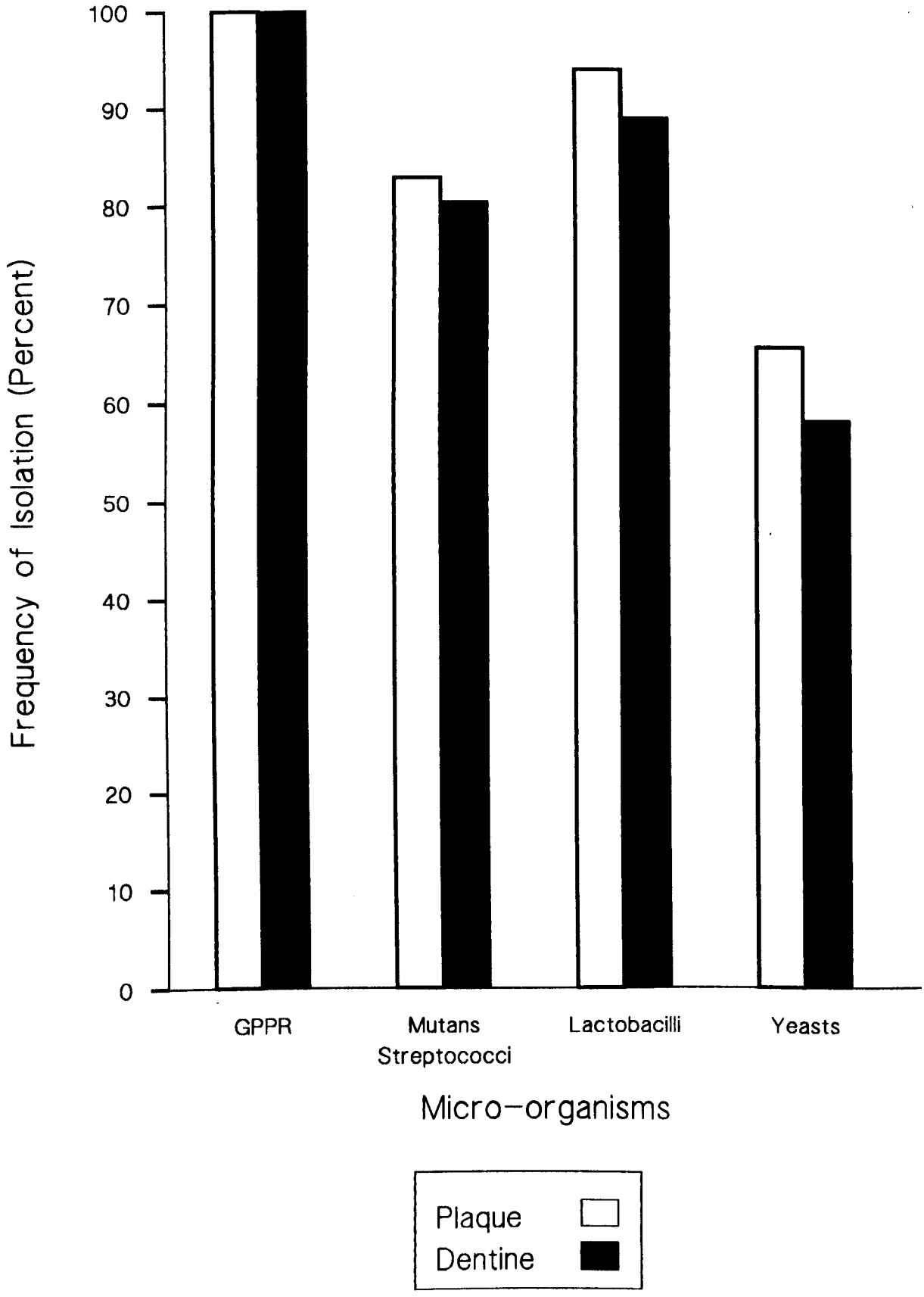
This histogram, displaying the percent of each of the four types of micro-organisms studied with reference to the total colony forming units cultured, clearly shows the dramatic differences between the Gram-positive pleomorphic rods in plaque (17.5 percent) and in dentine (36 percent), a significance of $P < 0.001$; between Lactobacilli in plaque (17.8 percent) and in dentine (14 percent), a significance of $P < 0.01$. The differences in the Mutans streptococci were not significant, ie in plaque (17.5 percent) and in dentine (14 percent).

V-703

The Frequency of Isolation of Dentine Micro-organisms

V

The Frequency of Isolation of Plaque Micro-organisms



V-703 The Frequency of Isolation of Dentine Micro-organisms v The Frequency of Isolation of Plaque Micro-organisms

The frequency with which all four taxa were isolated from either plaque or the underlying carious dentine were very high. Gram-positive pleomorphic rods were present in all samples taken; Mutans streptococci from about 80 percent; Lactobacilli from 90 percent or more and even Yeasts from about 60 percent. The differences within each taxon between plaque and dentine samples were not significant at the 0.05 probability level.

V-800 The Microbiology of Carious Dentine Subjected to Chemotherapy

This section presents the results obtained from 82 Primary Root Caries Lesions in 82 patients none of which were included in the investigations reported in V-600. They were selected for inclusion in these experiments as described in IV-312 and included 42 lesions diagnosed clinically as having Leathery Textures and with a Perceived Treatment Need to be restored and 40 lesions with a Soft Texture, also deemed to require restoration. On a random basis of selection, lesions from each of these groups were subjected to Chemotherapy as described in IV-313 whilst others received no Chemotherapy.

As with the microbiological data presented in V-600, the total numbers of colony forming units, Mutans streptococci, Lactobacilli and Yeasts were determined. However, the data reported in V-600 indicated that the Gram-positive pleomorphic rods, though the dominant organisms in all the biopsies cultured, were less significant between lesions with different characteristics, whilst the Mutans streptococci and Lactobacilli were more important variables when predicting Texture or Perceived Treatment Need. From the histograms displayed in V-400, the main significant variations between lesions were found in the numbers, proportions and frequency of isolation of Mutans streptococci and Lactobacilli rather than the Gram-positive pleomorphic rods. Therefore, the extensive procedures required to culture the Gram-positive pleomorphic rods were not undertaken for this series of experiments on the consequences of the topical application of a Chemotherapeutic agent.

The data are presented in two sections, the first (V-810) concerns the biopsies from Soft lesions, the second (V-820) with biopsies from Leathery lesions. In each section three histograms are presented, comparable to those in V-700, ie

- The Numbers of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy : V-811

- The Median Proportions of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy : V-812

- The Frequency of Isolation of Dentine Micro-organisms from Soft Lesions With and Without Chemotherapy : V-813

- The Numbers of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy : V-821

- The Median Proportions of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy : V-822

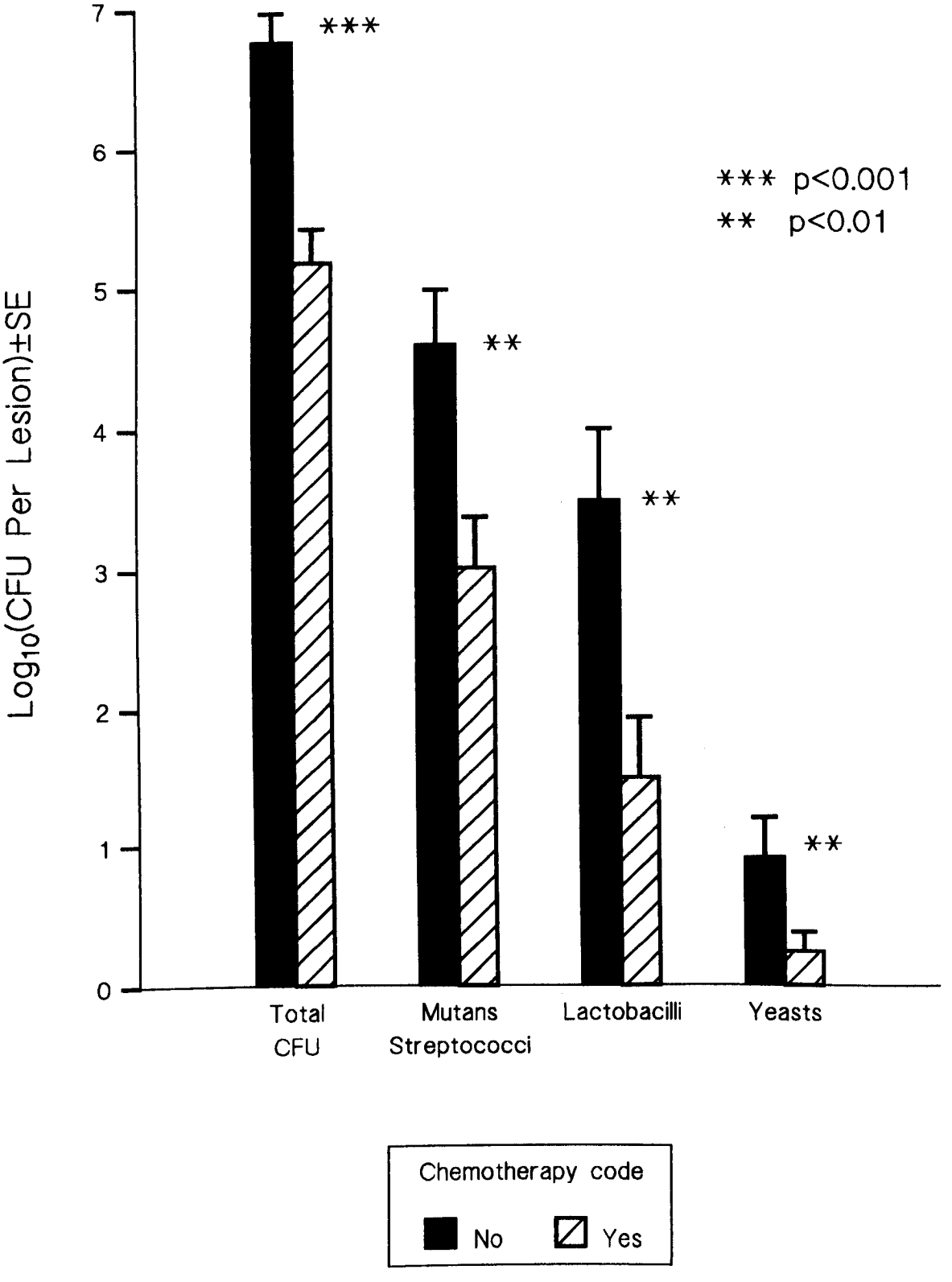
- The Frequency of Isolation of Dentine Micro-organisms from Leathery Lesions With and Without Chemotherapy : V-823

V-810 The Microbiology of Soft Lesions With and Without Chemotherapy

Of the 40 Soft lesions included in this experiment 20 were subjected to Chemotherapy and 20 were not subjected to Chemotherapy.

V-811

The Numbers of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy

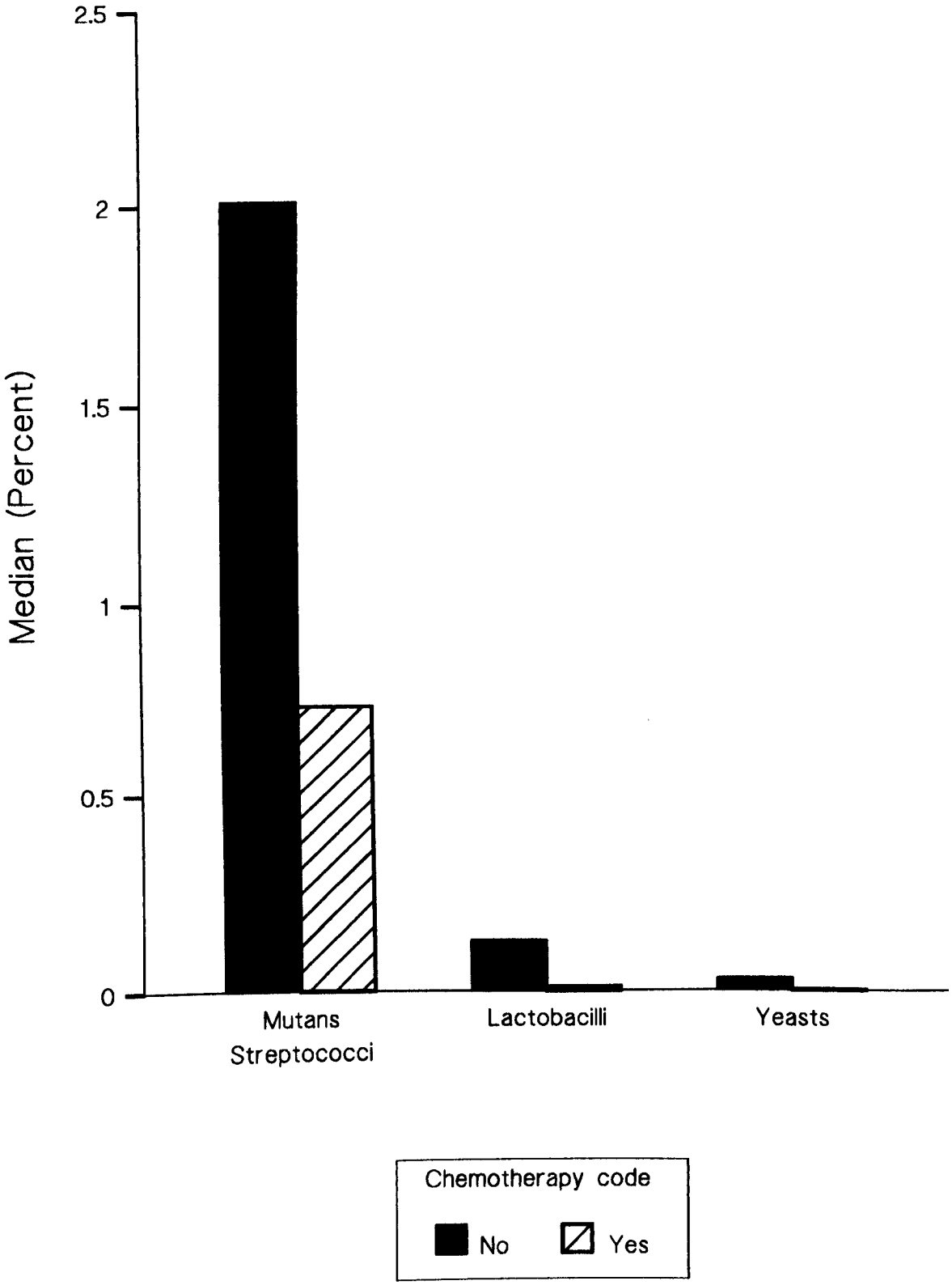


V-811 The Numbers of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy

Once again, this histogram, being a Log_{10} scale fails to illustrate the dramatic differences that were found between the microflora from the biopsies from lesions which received no Chemotherapy as compared to the microflora from the lesions treated by Chemotherapy. The total colony forming unit count (Mean ± Standard Error) in Soft caries was significantly reduced from Log_{10} 6.7 ± 0.2 to Log_{10} 5.20 ± 0.2, and this significance between groups was determined by using $P < 0.001$ confidence limits. Highly significant reductions were also found in the three individual counts : Mutans streptococci (Log_{10} 4.55 ± 0.4 to Log_{10} 3.00 ± 0.3); Lactobacilli (Log_{10} 3.50 ± 0.5 to Log_{10} 1.5 ± 0.4); and Yeasts (Log_{10} 0.90 ± 0.3 to Log_{10} 0.20 ± 0.2) ($P < 0.01$). It is also of value to note that the numbers of micro-organisms from these Soft lesions not subjected to Chemotherapy can be compared with those recorded in the larger sample of lesions reported in V-421 eg total colony forming units Log_{10} 6.7 ± 0.2 and Log_{10} 7.1 ± 0.05 respectively.

V-812

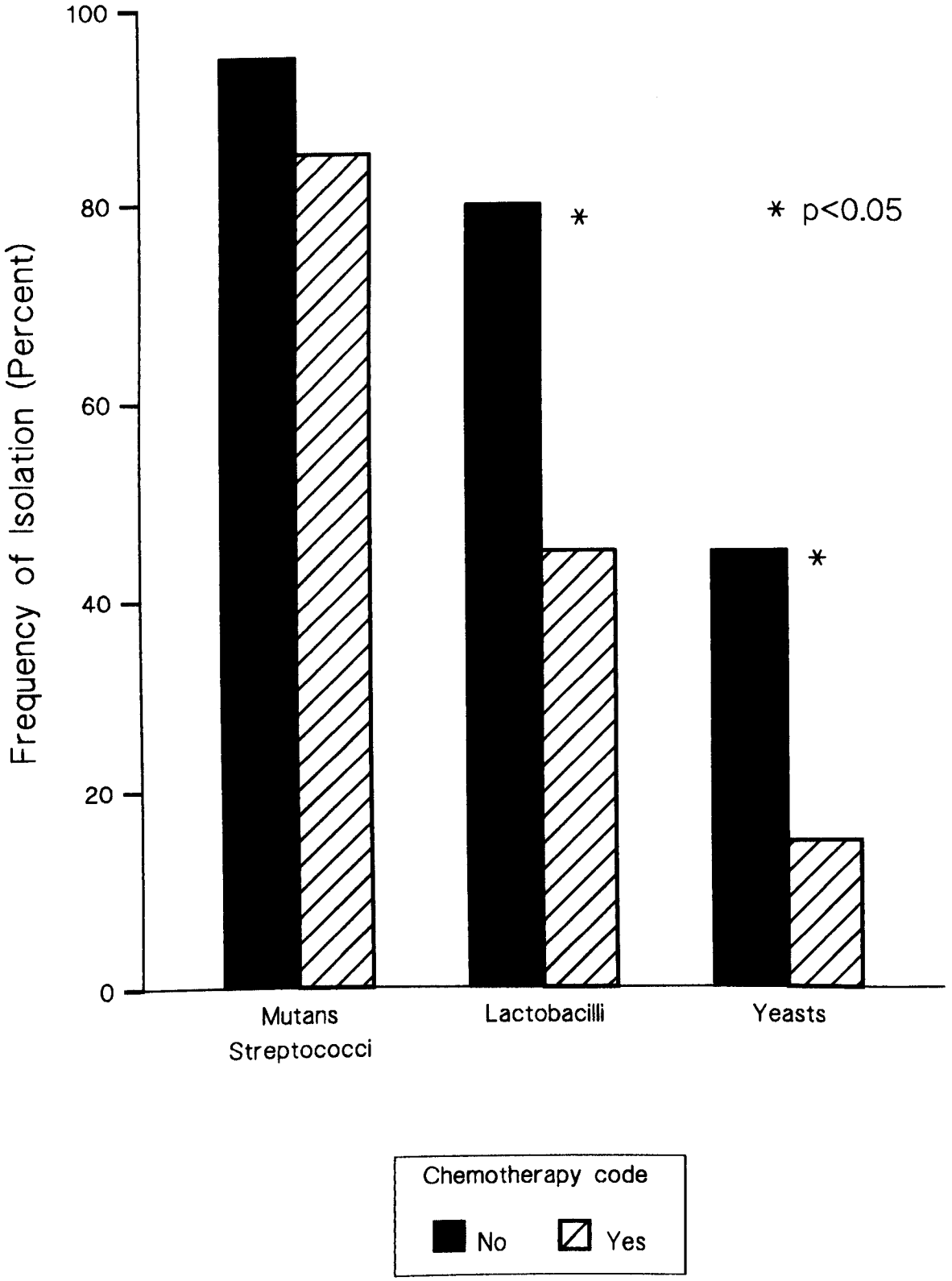
The Proportions of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy



V-812 The Median Proportions of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy

This histogram reveals that of the median proportions of the three taxa of organisms cultured from lesions which had not been subjected to Chemotherapy, Mutans streptococci (2.0 percent of all Colony forming units) were more than Lactobacilli (0.15 percent) and Yeasts (0.02 percent) ($P < 0.05$). In the Chemotherapy group these median proportions were reduced to 0.75 percent for the Mutans streptococci and to virtually zero for the other types. These reductions, however, were not significant within each taxon.

V-813
The Frequency of Isolation of Dentine Micro-organisms From Soft Lesions With and Without Chemotherapy



V-813 The Frequency of Isolation of Dentine Micro-organisms in Soft Lesions With and Without Chemotherapy

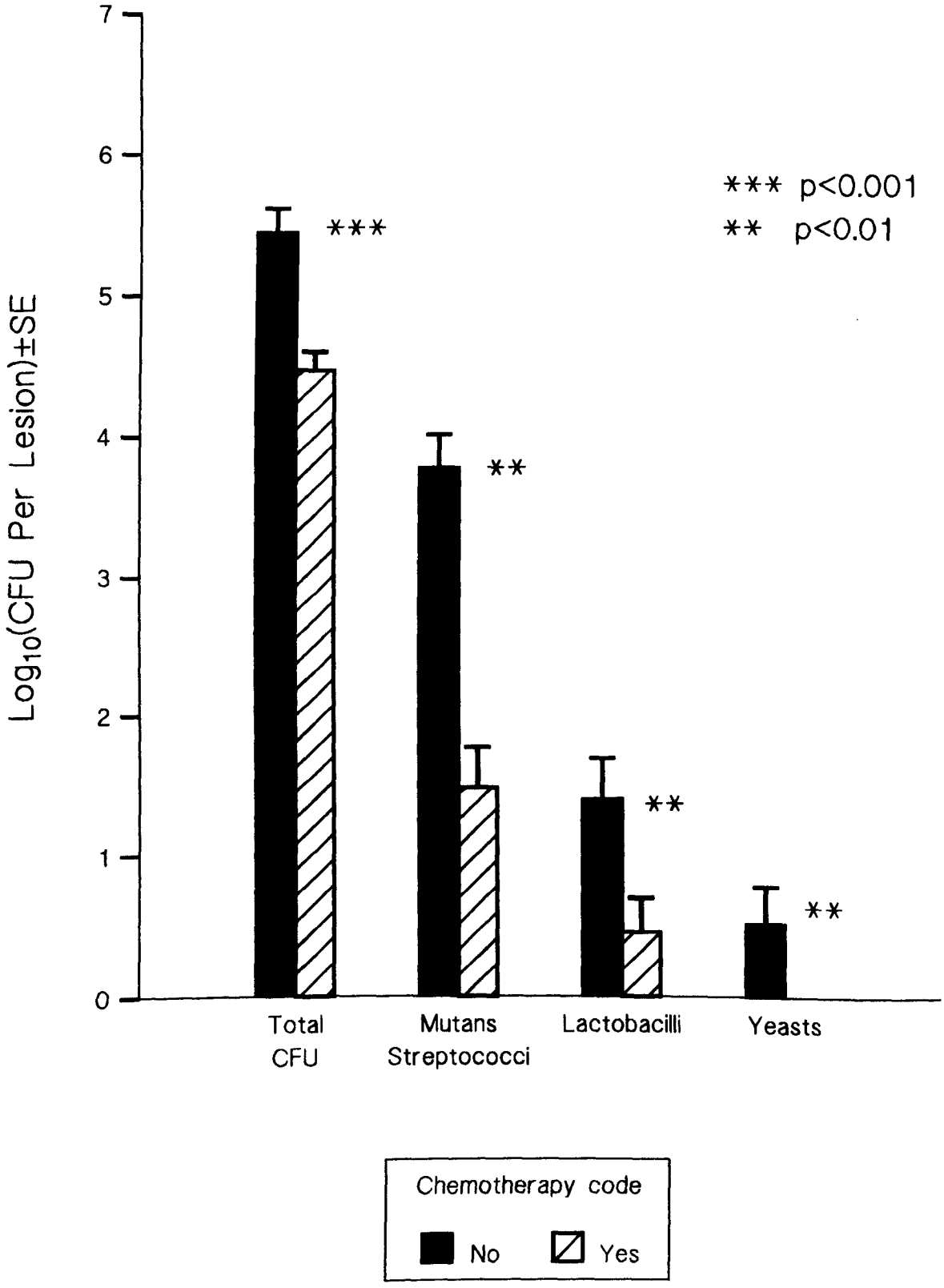
The Frequency of isolation of these three taxa from the Soft lesions not subjected to Chemotherapy can be compared with those recorded in the larger sample of lesions reported in V-423. Whilst the Mutans streptococci were isolated from 85 percent of Soft lesions which had been subjected to Chemotherapy, this reduction of 10 percent was not significant at the 0.05 probability level; whilst Lactobacilli were isolated from only 44 percent of lesions subjected to Chemotherapy, a reduction of 36 percent; and Yeasts from 18 percent of lesions subjected to Chemotherapy, a significant reduction of 26 percent. The significance between groups within these latter two taxa was determined by using $P < 0.05$.

V-820 The Microbiology of Leathery Lesions with and Without Chemotherapy

Of the 42 lesions included in this experiment, 22 were subjected to Chemotherapy and 20 were not subjected to Chemotherapy.

V-821

The Numbers of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy

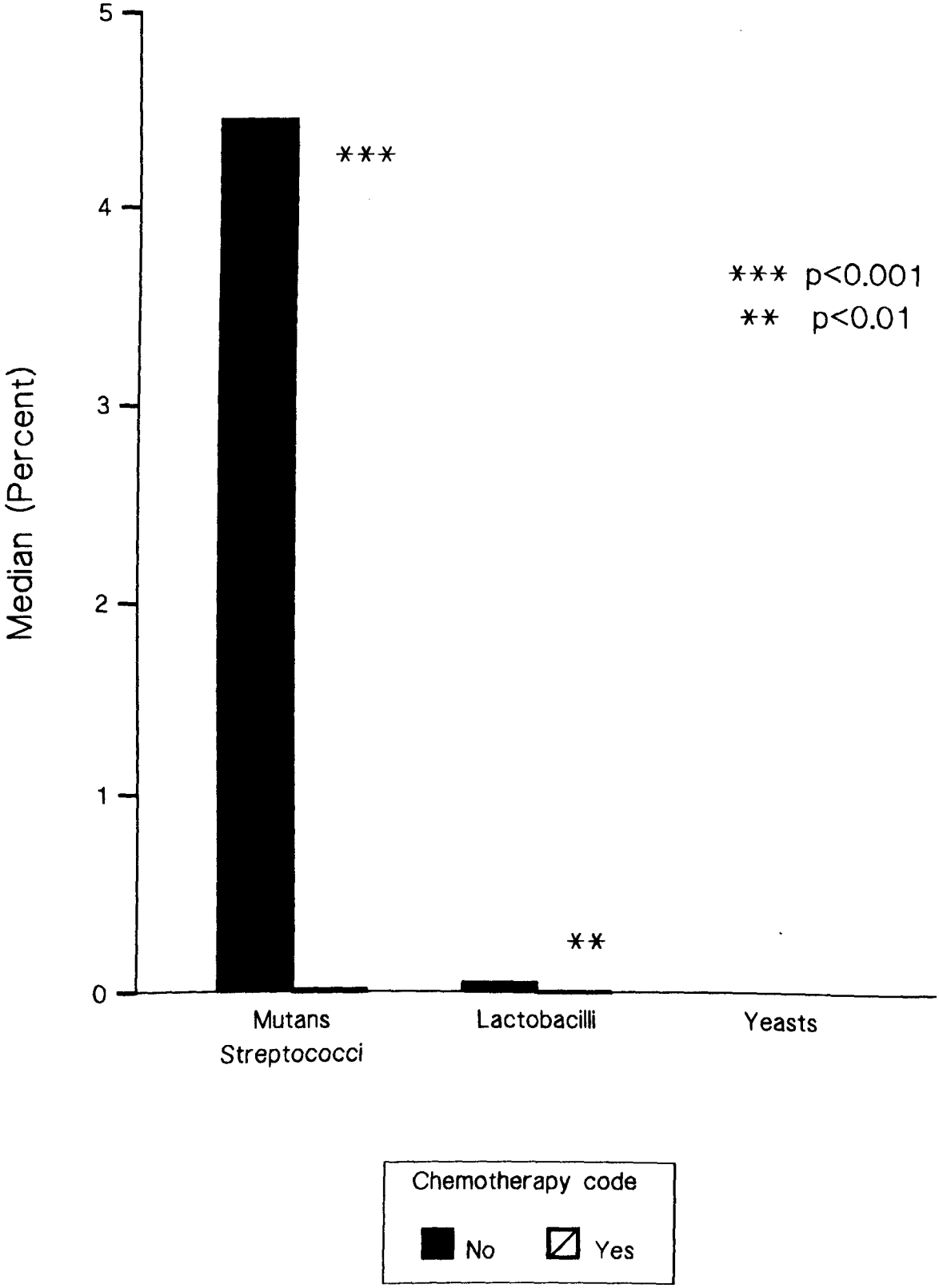


V-821 The Numbers of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy

These data on Leathery lesions mirror closely those presented in V-811 on Soft lesions. Though the total numbers are lower, the pattern of change is comparable and the significant levels of the changes produced are the same. The figures for untreated Leathery Lesions can also be compared to those obtained in the larger study recorded in V-421.

V-822

The Proportions of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy

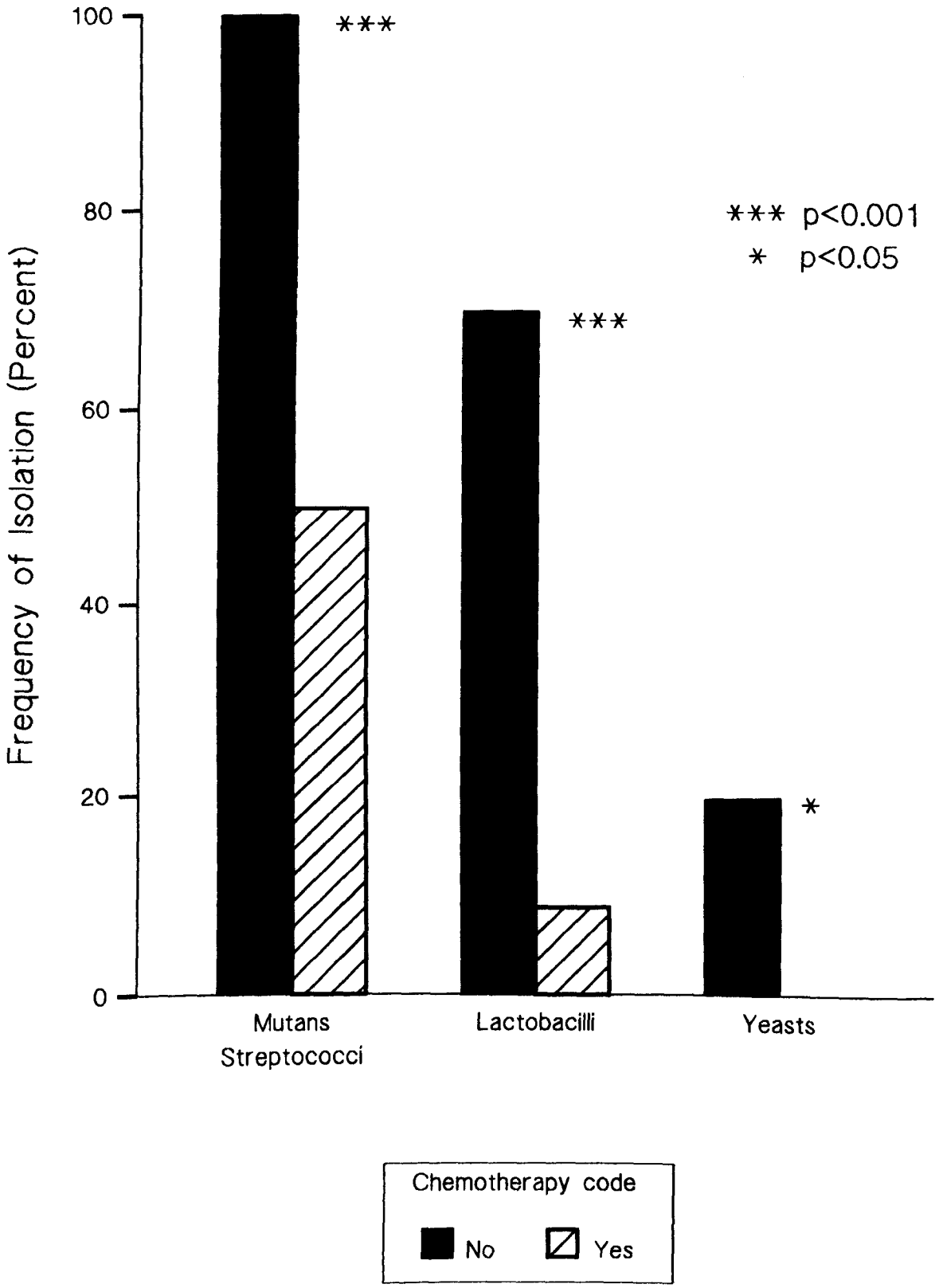


V-822 The Median Proportions of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy

The median proportions of Mutans streptococci were reduced from 4.4 percent from biopsies from Leathery lesions which had not been subjected to Chemotherapy to almost zero from the Leathery lesions which had been subjected to Chemotherapy, ($P < 0.001$), and the existing small median percent of Lactobacilli from 0.04 percent from Leathery lesions not subjected to Chemotherapy to 0.01 percent ($P < 0.01$).

V-823

The Frequency of Isolation of Dentine Micro-organisms From Leathery Lesions With and Without Chemotherapy



V-823 The Frequency of Isolation of Dentine Micro-organisms in Leathery Lesions With and Without Chemotherapy

The Frequency of isolation of Mutans streptococci was reduced from Leathery Primary Root Caries lesions which had not been subjected to Chemotherapy from 100 percent to 45 percent in Leathery lesions which had received Chemotherapy ($P < 0.001$); Lactobacilli from 70 percent to 9 percent ($P < 0.001$); and Yeasts from 20 percent to zero ($P < 0.05$).

VI DISCUSSION

VI-100 Introduction

Although Primary Root Caries is a significant clinical problem (Galan and Lynch 1993a) as discussed on II-130, the validation of the clinical criteria used to diagnose lesions has not been thoroughly addressed. Nyvad and Fejerskov (1987a) reported difference in the degree and pattern of mineralisation of lesions they classified as Soft, Leathery, or Hard. Soft lesions were characterised by extensive demineralisation with no evidence of an intact mineralised surface layer, while hard lesions appeared to have a generally uniform distribution of mineral throughout the lesion. They reported a broad range of histological appearances in Leathery lesions and concluded that Soft and Leathery lesions were active while Hard lesions were arrested. Studies reported by Schüpbach et al (1990a, b, 1992) confirmed these observations and reported bacterial ghosts within the remineralised tissue of Hard root caries. Histopathological destruction of root tissue is clearly associated with invasion of these tissues by micro-organisms. During this discussion, levels of caries activity, are discussed relative to the microflora isolated, ie active lesions are taken as those which have been clinically diagnosed with criteria deemed to be associated with 'active' demineralisation of tooth tissue and which contain significant acidogenic micro-organisms, whilst arrested lesions are taken as the opposite.

The core of this research has addressed certain microbiological characteristics of Primary Root Caries in relation to the clinical signs that such lesions present, in an effort to provide reliable evidence on which the levels of caries activity might be judged with some confidence, ie active or

arrested. Confidence in such judgements is fundamental for, on them treatment strategies, epidemiological data, and research into new philosophies of management are based. This discussion is presented under five headings:

VI-200 Variations of significance observed in the microflora of Primary Root Caries.

VI-300 Clinical signs as indicators of activity levels in Primary Root Caries.

VI-400 The Researcher's perception of Treatment Needs.

VI-500 The effects of topical Chemotherapy on bacterial activity in Primary Root Caries.

VI-600 Requirements for sampling Primary Root Caries for microbiological investigations.

VI-200 Variations of Significance Observed in the Microflora of Primary Root Caries

Microbiological identification is laborious, time-consuming, expensive and frequently restricted by the use of selective media which may exclude some potentially important species whilst inhibiting others being sought (Schaeken et al, 1986; Kimmel and Tinanoff, 1991); and even when non-selective media are employed, the detection limit implies that only the predominant microflora in a sample will be analysed. Nevertheless, microbiological examinations such as those that have been employed in this study are of considerable value and a number of legitimate conclusions can be drawn from them.

The histograms displayed in V-400 clearly demonstrate the enormous variations that were found in the number, proportions, and frequencies of isolation of Gram-positive pleomorphic rods, Mutans streptococci, Lactobacilli and Yeasts as well as all colony forming units from Primary Root Caries presenting with different clinical signs. However, though Gram-positive pleomorphic rods were invariably isolated in high proportions, frequencies of isolation, and total numbers, from lesions regardless of their clinical characteristics, it is clear that the main variations of significance between lesions are to be found in the numbers of Mutans streptococci and Lactobacilli (V-421, V422, V423 and V-600) which associate them with active caries processes. Other cross-sectional studies have implicated Mutans streptococci, either alone or in combination with Lactobacilli, as major pathogens of root caries because of their higher isolation frequency and/or their higher proportions in plaque on carious dentine surfaces when compared with that from sound surfaces (Billings et al, 1985; Brown et al, 1986; Fure et al, 1987; Keltjens et al, 1987a; Bowden et al, 1990; van Houte et al, 1990). However, only the present work is known to have identified these specific taxa within Primary Root Caries itself, having firstly excluded organisms present in any overlying plaque through its removal, and related them to the disease status determined from clinical signs.

Mutans streptococci and Lactobacilli are also, undoubtedly, important pathogens associated with enamel caries in humans (Hamada and Slade, 1980; Bowden et al, 1984; Loesche, 1986; Bowden, 1991) but, their presence does not necessarily indicate active disease is present, for Mutans streptococci have also been isolated from sound root surfaces (Brown et al, 1986; Keltjens et al, 1987a) and differences in the proportions

of Mutans streptococci have not been detected between sound and carious root surfaces (Ellen et al, 1985a, b,: Emilson et al, 1988). Such cross-sectional studies do not establish cause and effect relationships between particular bacterial species and the carious process. Longitudinal studies to monitor shifts in the microbial composition of plaque, over a period of time prior to the development of any clinical signs of caries, would be needed to determine such relationships.

Since caries undoubtedly has a microbiological origin and Primary Root Caries has varying levels of activity as discussed in II-240, it would seem to follow that caries, from which significantly higher levels of especially aciduric and acidogenic micro-organisms can be obtained, are those lesions which may be identified as being 'more active' than lesions with lower levels. The present work supports this assumption, notably that presented in V-421, V-422, V-423, V-601, V-602 and V-604. Even though Gram-positive pleomorphic rods predominate in all lesions, their numbers do not vary very much between lesions with different textures (V-422), therefore their numbers, however great, do not seem to provide a reliable indication of the level of 'carious activity'. For example, almost 30 percent of the microflora recovered from Hard, so-called arrested lesions, were Gram-positive pleomorphic rods (V-422).

Many microbiological studies have failed to show a positive correlation between *Actinomyces* spp and root caries (Billings et al, 1985; Ellen et al, 1985b; Brown et al, 1986; Fure et al, 1987; Keltjens et al, 1987a; Emilson et al, 1988; van Houte et al, 1990) and plaque from carious root surfaces tends to have lower levels of *Actinomyces* spp compared to plaque from sound root surfaces (Billings et al, 1985; Brown et al, 1986; Fure et al, 1987; van Houte et al, 1990). Bowden (1990) had also

suggested that heterogenous species or cariogenic biotypes may be hidden within the definition of *Actinomyces viscosus*. The work of Johnson et al (1990) reported that the taxonomy of *Actinomyces* spp is presently a dynamic one and that more research into this genus is required. The possibility, that amongst the numerous different species, collectively identified as Gram-positive pleomorphic rods, there are some which could be specifically implicated in the development of Primary Root Caries. Research is presently underway to determine whether or not this is the case. It is reasonable to conclude, therefore, that from these studies, the levels of *Actinomyces* spp in dental plaque do not provide reliable indicators of caries activity in underlying dentine. Their presence in plaque only emphasises the fact that Primary Root Caries exists in a complex and constantly changing oral environment in which a wide variety and number of micro-organisms exist. Furthermore, the microflora of Primary Root Caries is equally complex, variable and different from that of any related plaque.

Histological evidence has demonstrated how dentine is alternately decalcified, then re-calcified, with organisms eventually invading opened dentinal tubules as well as at the root surfaces, their progress beyond the tubules being inhibited for a time by the highly mineralised peri-tubular matrix before they progress through the less heavily calcified intertubular matrix. The bacterial invasion of the dentine occurs early in the carious process (Furseth and Johansen, 1970; Furseth, 1971; Jordan and Hammond, 1972; Frank et al, 1989; Schüpbach et al, 1989; Nyvad and Fejerskov, 1990), unlike enamel caries, presumably due to the structural differences between these two tissues. Clearly, the level of microbiological activity within a lesion will depend on the propensity of the immediate environment to demineralise the tissue before organisms of various types

can invade, therefore, the level of activity will depend on the ability of certain micro-organisms to lower the pH of the immediate environment. The most important organisms in this regard are apparently the Mutans streptococci and the Lactobacilli and the post-decalcification invasion, of other micro-organisms, notably the Gram-positive pleomorphic rods and the Yeasts, will depend on the lowering of the pH that has already occurred. Environmental pH plays an important regulatory role in the ecology of dentine caries. The low pH in a lesion (Dirksen et al, 1963; Fejerskov et al, 1992) provides an environment favoured by a limited number of acidogenic species, mainly Mutans streptococci and Lactobacilli (Loesche and Syed, 1973; Edwardsson, 1974) so Soft caries in which there has been extensive demineralisation equates with a relatively high rate of caries progression (Nyvad, 1993). The data presented throughout V-400 and V-600 supports this contention including the significance of Mutans streptococci and Lactobacilli in the occurrence of active Primary Root Caries. Nyvad (l993) also has reported that the presence of species within *Veillonella, Actinomyces, Streptococcus* and *Neisseria* have the ability to convert lactic acid into weaker acids which may dampen any caries activity, and alkali-producing species which may also participate in balancing acidity. Areas for further research include the relationships between the pH and the composition of the microflora of Primary Root Caries, and the overlying plaque, as well as the clinical criteria used to diagnose lesions.

Mutans streptococci have a high cariogenic potential possibly due to a unique combination of properties which these micro-organisms possess (Hamada and Slade, 1980; Tanzer, 1981; Bowden et al, 1984; Loesche, 1986; Hamada et al, 1986; Lindquist et al, 1989; Carlsson, 1989; and Bowden, 1991). They have the capacity to adhere to, and accumulate on,

the surfaces of teeth due to their production of extracellular water-insoluble polysaccharides from sucrose; they are highly acidogenic and aciduric and synthesise intracellular polysaccharides from carbohydrates; they are able to metabolise several of the salivary glycoproteins which are adsorbed to the tooth surface and, they also produce acid phosphatase which can hydrolyse the tooth matrix and then translocate phosphorus from the tooth into their own bacterial cells. Other oral micro-organisms have some of these properties, but since Mutans streptococci have them all, they are highly significant to the carious process. However, other bacteria, or combinations of other acidogenic or aciduric micro-organisms, will undoubtedly, also contribute to the pathogenesis of Primary Root Caries, and their potential will be highly dependent on numerous environmental factors. Amongst these is the fact that highly acidogenic and aciduric Lactobacilli which depend on dietary carbohydrate for colonisation and colonise areas of stagnation, produce acidic environments which result in the progression of Primary Root Caries. The results reported in V-400 confirm the dominant roles of both Mutans streptococci and Lactobacilli in the occurrence of the Primary Root Caries process.

The cariogenic potential of the Mutans streptococci and Lactobacilli ties in with the role of dietary factors as discussed in II-221. Frequent consumption of carbohydrates, conducive to development of the caries process, as well as the emergence of Mutans streptococci and Lactobacilli, may allow these highly acid-tolerant organisms to have a selective advantage over most of the other less acid-tolerant microflora when the plaque milieu is frequently acidified (Denepitiya and Kleinberg, 1984; van Houte and Russo, 1986). Therefore, the essential condition for the emergence of the Mutans streptococci and Lactobacilli is not the dietary

sucrose or carbohydrate availability *per se*, but, instead, the frequency of recurrence of acidic episodes in dental plaque that accompany frequent carbohydrate consumption, or even, perhaps, the frequent ingestion of acidic foods and drinks. Research to investigate this concept, *in vivo* is being planned. Some support for this concept has already been published (Svanberg, 1980; Bradshaw et al, 1989, van Houte, 1993). This lowering of the pH, in turn, increases the probability of a net loss of soft tissue minerals over time.

The level of Yeasts in whole stimulated saliva is significantly correlated with the number of active Primary Root Caries lesions and with the total numbers of decayed and filled root surfaces in an elderly population by Beighton et al (1991c), and similar observations have been made regarding the association between coronal caries and levels of Yeasts in the saliva of children (Peinihakkinen et al, 1987). In this present study Yeasts were more frequently isolated and in the greatest numbers from lesions nearest to the gingival margin (V-433); from Soft lesions (V-423); and from lesions deemed to require restoration (V-503). These associations would seem to reflect the acidogenicity and aciduricity of the organisms, since lesions at the gingival margin are more likely to be Soft (V-221) and require restoration (V-303) and, therefore, to be active, with micro-organisms penetrating into the dentinal tubules producing dentine demineralisation and proteolytic destruction of the dentinal organic matrix (Schüpbach et al, 1990b). The mean numbers of Yeasts isolated from Leathery and Hard lesions were significantly different from each other, whilst both mean values were significantly less than that associated with Soft lesions (V-421). Given the pattern of distribution of Soft, Leathery and Hard lesions, with respect to their distance from the gingival margin (V-221),

the differences in Yeast numbers may reflect differences in the activity of Primary Root Caries. Hard lesions were probably once Soft, passed through a Leathery stage and, upon gaining an increased distance from the gingival margin, become remineralised and Hard. However, longitudinal clinical trials would be required to investigate such an hypothesis. Associated with these changes in mineralisation may be these changes in yeast levels (V-421) and their Frequency of isolation (V-423). It would seem unlikely that Yeasts play an important and active role in the progression of Primary Root Caries as their numbers are far lower than the total number of bacteria within Soft lesions (V-421), where they are not always found (V-423). Perhaps, it is more appropriate to regard them as a marker organism which, when present, is indicative of active Primary Root Caries (Beighton and Lynch, 1993b).

The closeness of the associations between variables is shown in V-601, which again underlines the importance of the Mutans streptococci and the Lactobacilli as risk factors for Primary Root Caries. Interpretations of the multiple regression analyses in V-604 and V-607 and the discriminant analyses in V-602, V-603, V-605 and V-606 further reinforce the influence of these micro-organisms on the level of activity in such lesions. However, the independent variables in multiple regression analyses may be interrelated, so the results must be interpreted with caution.

It would seem to be reasonable, therefore, to conclude that the numbers, proportions and frequency of isolation of the Mutans streptococci and/or the Lactobacilli provide the most reliable microbiological indications of the levels of activity within Primary Root Caries, ie the greater these figures for these two organisms, the higher the level of carious activity in the lesion.

VI-300 Clinical Signs as Indicators of Activity Levels in Primary Root Caries

Microbiological investigations, such as those employed in this study, cannot reasonably be employed by clinicians on a day to day basis to determine the most appropriate treatment strategy, nor can they be used by epidemiologists in their data collection. It is, therefore, necessary to relate those data which are available, ie the clinical signs of Primary Root Caries, to microbiological data in order to establish confidence in what these signs indicate.

Clinicians tend to equate 'stained' dentine with 'carious' dentine and assume that carious lesions should, ideally, be made stain-free before restoration, though an exception is made with respect to circumpulpal dentine exposed in deep lesions. However, it is suggested that stained circumpulpal dentine is not removed more because this would probably result in the actual exposure of the pulp rather than in any absolute conviction that it is inactive. The Colour of root dentine has, therefore, generally been accepted not only as indicating the presence or absence of caries, but also whether or not it is an active lesion and is likely to progress. It has been customary for many clinicians and researchers (Banting and Ellen, 1976; Nyvad and Fejerskov, 1982; Nordenram et al, 1988; De Paola et al, 1989a; Fejerskov et al, 1991; Todd and Lader, 1991; Adriaens and Nyvad, 1992; Eliasson et al, 1992) to assume that the darker a lesion, the more likely it is that the carious process is arrested or only slowly progressive, whilst light coloured caries has generally been considered to be active (Banting and Ellen, 1976; Hix and O'Leary, 1976; Nyvad and Fejerskov, 1982, 1986; Miller et al, 1987; Nordenram et al, 1988; De Paola

et al, 1989a; Fejerskov et al, 1991; Todd and Lader, 1991; Ravald and Birkhed, 1991; Adriaens and Nyvad, 1992; Eliasson et al, 1992). The present work shows that such views are probably in error and too simplistic (Lynch and Beighton, 1994).

The three histograms in V-410 present the numbers of micro-organisms obtained from the three Colours of lesions diagnosed, in the descending order: Black; Yellow; Light Brown; then Dark Brown. Whilst the latter three categories do indeed follow the commonly assumed order indicating that more active lesions are light coloured and less active ones darker, the marked exceptions are the Black lesions. Rather than being revealed as the least active group of lesions, they are shown to be the most active and especially so with respect to the Mutans streptococci and Lactobacilli counts (V-411, V-412 and V-413). This is unfortunate since clinically it is not always easy to distinguish between the least active lesions (Dark Brown) and the most active lesions (Black) and this must introduce significant doubt into epidemiological data which depends so heavily on visual appearance, as discussed in II-130. The present study, and an earlier one (Hellyer et al, 1990), support Kidd et al (1993) who reported the inability of Colour to differentiate the level of infection in secondary caries diagnosed at enamel-dentine junctions.

In view of this uncertainty regarding the Colours of lesion, which clinical signs are shown to have significant correlations with caries activity as indicated by the levels of Mutans streptococci and Lactobacilli? Studies of the Locations of Lesions (V-431, V-432, V-433, V-601, V-602, V-603, V-604, V-605, V-606 and V-607) are especially interesting, for they show that the nearer a lesion is to the gingival margin, the more likely it is to be

actively carious, whilst lesions more remote from that margin tend to be less active. Relating Location to Colour (V-212, V-411, V-412 and V-413) Black lesions are found in significantly higher proportions when located less than 1 mm from the gingival margin than amongst those more distant from the soft tissues, ie a dark lesion < 1 mm from the gingivae is significantly more likely to be Black, with high counts of Lactobacilli and Mutans streptococci and therefore active, than a dark lesion > 1 mm from the gingivae, which is significantly more likely to be Dark Brown with lower counts of these two types of organisms and, therefore, relatively less active. Similar conclusions can be drawn with respect to Yellow and Light Brown lesions.

It is, perhaps, not surprising that the most active lesions tend to occur close to the gingival margin, for in such Locations they tend to be more protected from mechanical disturbance, through the mastication of food or from toothbrushing, which permits the stable environment in which plaque may exist undisturbed. These regions immediately above the gingival margin on exposed root surfaces are therefore stagnation sites which are likely to be preferentially colonised by acidogenic and aciduric micro-organisms with the potential to initiate and maintain the progression of Primary Root Caries. After thorough tooth cleaning, any initial colonisation of root surfaces by micro-organisms occur primarily as colony-like flecks on retentive areas along the gingival margins (Björn and Carlsson, 1964; Bergström, 1981), being assisted by cracks or other surface irregularities (Moreau, 1984; Quirynen and van Steenberghe, 1989). A narrow plaque-free zone of root surface has been claimed to separate such deposits from the gingival margins themselves (Frank and Houven, 1970; Bergström, 1981; Quirynen et al, 1985), for crevicular fluid has

antimicrobial properties, but plaque then spreads in a coronal direction. With the recession of the gingivae and the resultant potentially greater exposure to mechanical cleansing of any gingivally located lesion it is to be expected that the level of carious activity would reduce.

As Primary Root Caries becomes more distant from the gingival margin, the balance of activity tends, overall, to favour remineralisation rather than demineralisation. It is probable, therefore, that Soft lesions evolve through the stages diagnosed as Leathery then Hard as the distance from the gingival margin increases. In support of this view, similar changes in the clinical severity of active root caries lesions have been reported as a consequence of: improved oral hygiene and the use of a fluoridated dentifrice (Nyvad and Fejerskov, 1986; De Paola, 1993); the application of fluoride or chlorhexidine varnishes (Schaeken et al, 1991a); or intensive oral hygiene instruction, fluoride therapy and professional tooth cleaning (Emilson et al, 1993).

It would be both interesting and advantageous to supplement the present work, employing techniques which would precisely identify the Location, especially with reference to the gingival margin as well as the Size of each dentine biopsy removed, and indicate plaque distribution and thickness related to each one, but to date, no study has systematically quantified the macroscopic pattern of microbial colonisation on exposed root surfaces. Such work is now planned and will involve the replication of plaque, tooth and gingival surfaces before and after sampling; recording co-ordinate data at 10 micron intervals over the surfaces of these replicas; and superimposing these data to determine with great precision the Size and relationships of biopsies and plaque thicknesses.

To supplement the clinical assessments of Colour and Location it has proved possible by simple observation, rather than investigation, to estimate the Size of a lesion (V-214) and the degree of Cavitation (V-213). In general, the larger lesions tend to be Black or Yellow, ie in the two categories which have been identified with high levels of bacterial activity, whilst smaller lesions tend to be in the less active Colour groups (V-451, V452 and V-453). It can be argued that this is to be expected for, if microbiological activity is high then it is most likely to result in extensive destruction and in extension of the lesion, ie many such lesions would be large. The degree of Cavitation of a lesion is less clearly associated with its Colour and, therefore, with its microbiological activity (V-441, V442 and V-443). This is perhaps, again, not surprising since any subjective judgement of depth or loss of tissue, from what are usually markedly curved surfaces is notoriously difficult, and the causes of surface loss are multifactorial. More accessible surfaces are more likely to be subject to tissue loss from abrasion; more convex surfaces will lose more hard tissue than flat or concave surfaces; and inter dental lesions less than others.

Using only observable clinical signs of Primary Root Caries, this work suggests that epidemiologists in particular, who are generally not free to institute any investigations such as assessments of Texture, should take special care to differentiate between Dark Brown and Black lesions when categorising Colour of Primary Root Caries and should also take particular note of the Locations and Sizes of lesions when diagnosing them as active, inactive, or arrested.

However, when dentists examine lesions they firstly identify them visually but they are free to 'Investigate' each lesion, by means, other than pure visual ones before determining the most appropriate treatment strategy to advise and undertake. Instrumentation helps to define the limits of a lesion and the surface contour of an affected root surface but, most of all it, permits a subjective clinical assessment of the Texture of the affected dentine to be made. The data presented in V-420 and V-600 is very revealing and it is believed to demonstrate that Texture is the most reliable of all clinical predictors of microbiological activity in Primary Root Caries.

Three categories of Texture have been defined: Soft, Leathery, and Hard. Each group of data presented in V-420 quite clearly identifies Soft lesions as the most active and Hard lesions as the least active. Virtually no Mutans streptococci and no Lactobacilli were found in Hard lesions whilst Soft and Leathery lesions contained significant numbers. It is not surprising that Lactobacilli were not found on Hard Primary Root Caries for in comparison with other Textures, they are not in plaque retentive locations and the rise in the numbers of Lactobacilli in plaque overlying carious lesions may not reach a high level until after an active lesion can be detected clinically (Ikeda et al, 1973; Brown et al, 1975; Loesche et al, 1984; Macpherson et al, 1990). In addition, the surfaces of every Primary Root Caries lesion in this study was smooth and shiny on each occasion and this was not as conducive for plaque retention, as, for example, the rough surface of every Soft lesion. The preponderance of Gram-positive pleomorphic rods in all lesions is countered by the fact that the differences between the three Texture groups are less than occurs with Mutans streptococci or Lactobacilli, and the two groups of micro-organisms believed

to be most closely associated with active caries found only in Soft and Leathery lesions. Significantly different numbers of micro-organisms have been recovered from lesions with different Textures, which may be related to their states of mineralisation, as has been observed in electron microscopy studies (Nyvad and Fejerskov, 1987a; Schüpbach et al, 1990a, b, 1992). Some of the micro-organisms recovered from samples of Hard lesions might be those adhering to the surface of the altered root dentine even after surface cleansing. From the Leathery lesions, the micro-organisms recovered would seem to be those actually infecting the carious root dentine but, depending on the clinical severity of individual lesions, they may not necessarily be involved in the active destruction of root dentine. Soft lesions undoubtedly indicate an active Primary Root Caries process whilst Hard Primary Root Caries, regardless of its Colour or other characteristics, does not require intervention on the basis of its activity. It may well be appropriate to restore such a lesion for reasons of aesthetics or simply to restore surface morphology following extensive loss of dentine, but not because of an active microbiological state. It is believed that clinicians should clearly understand this, not least in view of the long standing belief of the overriding significance of the Colour of a lesion.

The association between the Textures and the Colours of lesions is remarkable (V-211), for Soft lesions (ie active) are predominantly Black or Yellow, the very Colour characteristics of more active caries, whilst Light Brown and Dark Brown lesions are more commonly Leathery or Hard. Similarly, Soft lesions are mostly at the gingival margin (V-221) whilst Hard lesions are found more remote from it. Perhaps, not surprisingly, Hard lesions tend to be less cavitated than Leathery lesions which are in their

turn, shallower than Soft lesions (V-222). Presumably, Soft lesions are more easily abraded and eroded than Hard lesions and, therefore, tend to loose more dentine from their surfaces. There are similar associations (V-223) between the Textures and the Sizes of lesions, for Soft lesions, which have been shown to be active, are most likely to be large and these too are shown as being active in their own right. Similarly, small and Hard lesions are relatively inactive.

Without doubt, the most certain diagnostic characteristic of which clinicians can make use in order to assess the microbiological activity likely to be present in any lesion, is to judge its Texture. Though this criterion alone is of little help to epidemiologists, its highly significant relationship, not only to the microbiological characteristics of lesions, but also to their clinical signs, provides evidence which confirms some assumptions that have been made on the levels of caries activity whilst denying others which are believed to be misleading. The Texture of caries is also a characteristic with which dentists are very familiar, for they daily use their judgement based on textural differences, to identify and remove calculus, to 'plane' root surfaces as well as to differentially diagnose sound and carious dentine. It will not be a surprise to any experienced dentist that large, Soft lesions located at the gingival margin are likely to be active, what may not have been appreciated to the same extent is that Black lesions are the most active of all the Colour groups defined for Primary Root Caries.

VI-400 The Researcher's Perception of Treatment Needs

The Perceived Treatment Needs as described in this work cannot be considered as every dentist's perception of the Perceived Treatment Needs of their own patients. They are far more limited than that, for they are only the researcher's perceptions of treatment needs and it is recognised that, as this work has proceeded, there can be no guarantee that the perceptions of need would not be affected by increasing knowledge accruing from the work itself. Nevertheless it is believed that the results recorded in V-300, V-500 and V-600 are valuable.

Strictly speaking, four categories of management for the diagnosed Primary Root Caries studied were defined, over and above plaque and dietary control:

- no treatment prescribed;
- only the topical application of a Chemotherapeutic agent instituted;
- the dentine diagnosed clinically as being carious was removed but no other action was taken;
- the dentine diagnosed clinically as being carious was removed and the resultant caries free lesion restored.

However, the last group has been divided into two on the basis of the Texture of the caries (Soft or Leathery). The three histograms in V-500 appear to demonstrate that the strategies selected for lesions on the basis of their clinically observed characteristics are in line with the degrees of activity within the lesions as determined through microbiological analysis.

Thus, few Mutans streptococci or Lactobacilli were found in the lesions for which no treatment was prescribed whilst the presence of these organisms became progressively more evident as more and more radical intervention was thought to be appropriate. There would seem to be some comfort in these findings, in that careful consideration of the clinical characteristics of Primary Root Caries can result in treatment strategies, which are quite compatible with decisions that would have been made had the microbiology of each lesion been known. Thus, the associations that have been discussed above, regarding the level of activity in lesions, would seem to be confirmed in respect of the treatment strategies prescribed, and their suitability in respect of the microbiological activity to the affected dentine.

Two different clinical methods have been used for the examination and classification of Primary Root Caries lesions. The first uses the criteria outlined by Nyvad and Fejerskov (1987c), and V-420 provides considerable microbiological data that are in accord with their histological observations of each of these lesion types, for they concluded that Soft and Leathery lesions were actively carious and Hard lesions were inactive. The microbiological results in V-500 suggest that this is an over-simplification which might be satisfactorily overcome by assigning Perceived Treatment Needs to individual lesions, on the basis of which a more appropriate classification might be made than that of Nyvad and Fejerskov (1987c), in which active and inactive lesions were based on lesion texture. Some of the present work has been published incorporating this suggestion (Lynch and Beighton, 1993, Beighton et al, 1993a). V-500 shows that Mutans streptococci and Lactobacilli are present in greater numbers and greater proportions, and are more frequently isolated from lesions with greater

Perceived Treatment Needs than from lesions requiring none, or minimal treatment. Root lesions designated for restoration were more likely to be Black or Yellow (V-301), to be more cavitated (V-304), to be larger (V-305) and to be closer to the gingival margin (V-303). These latter points may reflect the fact that Mutans streptococci and Lactobacilli are favoured by an environment which is conducive to caries. However, it should be noted that some lesions to be restored did not contain Mutans streptococci and Lactobacilli which underlines the complexity of the microbial community and that many other factors are involved.

VI-500 The Effects of Topical Chemotherapy on Microbial Activity in Primary Root Caries

Many strategies have been suggested for the treatment of the root caries process including professional prophylaxis (Banting et al, 1985); various fluoride programs (Billlings et al, 1985; Ögaard et al, 1990; Ravald and Birkhed, 1992); intensive oral hygiene procedures with a fluoridated dentifrice (Nyvad and Fejerskov, 1986; De Paola, 1993); fluoride or chlorhexidine varnish treatment (Schaeken et al, 1991a); intensive oral hygiene therapy, fluoride therapy and professional tooth cleaning (Emilson et al, 1993); intensive oral hygiene, daily self-application of fluoride and periodic professional application of fluoride (De Paola, 1993). However, no study has previously been undertaken to test the antimicrobial effects of a Chemotherapeutic agent by obtaining biopsies of carious root dentine after the removal of any overlying plaque and analysing it microbiologically.

The data presented in V-800 is in fact more dramatic than perhaps the histograms suggest, for the numbers of micro-organisms are by necessity displayed on a Log_{10} scale. Statistically, the suppression of microbiological activity in the carious dentine treated Chemotherapeutically is significant and all the 82 lesions from 82 patients entered in this study were diagnosed as deemed to require a restoration, so the potential consequence of these observations on treatment strategies for Primary Root Caries may be very great indeed. The presence of active caries, as already discussed, is associated with high recoveries of acidogenic and aciduric micro-organisms such as Mutans streptococci and Lactobacilli (Ellen et al, 1985a, b; Brown et al, 1986; Keltjens et al, 1987a; Emilson et al, 1988; Bowden et al, 1990; van Houte et al, 1990; Nyvad and Kilian,

1990a), whilst a more complex microflora has been found in the plaque from root surfaces exhibiting low caries activity (Nyvad and Kilian, 1990b). Despite a limited number of subjects (6) and of lesions examined (19), Nyvad and Kilian (1990a) claimed that the highest mineral loss in Primary Root Caries is associated with a domination of acidogenic species such as Mutans streptococci and Lactobacilli or is characterised by a complex microflora, including Mutans streptococci and Lactobacilli. V-800 clearly shows that Mutans streptococci and Lactobacilli, as well as the total colony forming units and Yeasts are reduced in lesions treated Chemotherapeutically.

Note has already been made of the clinical problems associated with the traditional strategy of surgically removing carious dentine and restoring the affected root, ie access, isolation, trauma, matricing, and finishing. If a quite different strategy could be proven as effective, involving the conservation of the carious dentine, its sterilisation or inactivation, and the maintenance of this condition, then the prognosis for teeth affected by the Primary Root Caries process, the continuing costs of their maintenance and all the emotional and psychological consequences of the surgical management of the disease, would disappear. Furthermore, if such Chemotherapeutic strategies were to be proven as appropriate for the management of Primary Root Caries, it is difficult to believe that comparable approaches would not prove to be appropriate for the management of caries of coronal dentine in both children and adults with arguably even greater overall advantages. However, the investigations reported here are but a preliminary study. The carious dentine was not sterilised, though the colonisation by micro-organisms was dramatically

reduced. Nor is any evidence presented as to for how long the observed effect might be maintained. The differences observed of the effects on Soft and Leathery lesions respectively serve as a warning that far more extensive investigations are indicated before sound conclusions might be drawn and confidence felt in any possible recommendations that might come to be made. The use of the research techniques mentioned in VI-300 to identify the precise Location and Size of the dentine biopsy will also be advantageous to supplement this work. For example, one may speculate that Soft lesions are deeper than Leathery lesions, from the outer surface of the caries to the inner surface of the infected tissue, and that topical Chemotherapy is unable to penetrate to the full depths of the Soft lesions. On the other hand, these results were achieved after only five applications of the agent over a period of less than one minute and no adverse effects were encountered. Patients reported no deleterious side effects which included soreness, odour, taste and ability to taste salt (Long et al, 1977). During the oral examination stage no additional staining of the teeth and no desquamation of any oral soft tissues were observed. It is also noteworthy that no varnish remained visible on the surface of any of the lesions after brushing and before sampling.

Investigations are already underway in the form of more extensive longitudinal studies of this alternative Chemotherapeutic treatment strategy which, it is hoped, will provide further evidence of its efficacy. It is also appreciated that investigations need to be made regarding the most effective and safe concentrations of the two agents: chlorhexidine and thymol; the potential value of alternative anti-microbial agents; the value of the 86 percent ethanol media used and any possible alternatives; the

effects following use of a placebo varnish; the advantages and disadvantages of incorporating varnish into the solution, including the consequences to the possible remineralisation of the carious dentine which it is well known can occur within the oral environment. The potential is fascinating and of considerable significance but much sound investigatory work is needed to determine just what is possible.

VI-600 Requirements for Sampling Primary Root Caries for Microbiological Investigations

The consistency of the sampling procedure and of the recording of the clinical signs and Perceived Treatment Needs of lesions is reflected in the low variance associated with the numbers of micro-organisms recovered from lesions of different clinical severity. Because the procedures used for the preparation of root surfaces before biopsying carious dentine removed all the extraneous plaque overlying the lesion, these samples more reliably reflected the micro-organisms associated with the lesions themselves than samples in many other studies, and more confidence can be felt in the inter-relationships that have been identified between the micro-organisms recovered from lesions and the classification of the lesions by clinical signs or by any clinical index based on Perceived Treatment Needs.

It is clear from the data presented in V-700 that, though plaque accumulating on the surfaces of roots together with the availability of readily fermentable carbohydrates for the maintenance of the microflora in it, is the major element initiating Primary Root Caries, the microbiology of plaque on the one hand, and of dentine caries on the other, differ considerably (Beighton and Lynch, 1994). Clearly, this reveals a need for high precision in any sampling technique, if reliable microbiological data is to be generated, notably plaque samples must be distinguished and separated from samples of carious dentine or a confused picture will be created. For these reasons every effort has been made in these current studies to eliminate plaque from any dentine samples obtained.

One may speculate why the proportions of Gram-positive pleomorphic rods are greater in active lesions than in the overlying plaque (V-702). Gram-positive pleomorphic rods tend to invade bundles of collagen fibres on root surfaces (Nyvad, 1993) in contrast to some other members of the plaque microflora (Nyvad and Fejerskov, 1987c). *Actinomyces* spp are not considered to be very proteolytic (Slack and Gerencser, 1975), however, selective binding to collagen may be a factor that promotes the invasion by *Actinomyces* spp into collagenous tissues (Liu et al, 1991) and Actinomyces-like bacteria selectively invade demineralised root tissue (Nyvad and Fejerskov, 1990). In addition, nutritional factors may favour preferential colonisation by *Actinomyces* in carious dentine as compared to the colonisation of the overlying plaque. *Actinomyces* spp may have a selective advantage over other common plaque micro-organisms such as Streptococci because of their more efficient utilisation of organic material if the environment is limited in carbohydrate (van der Hoeven et al, 1984; Rogers et al, 1986); and as suggested by the lower acid production after a sucrose challenge in the inner layers of plaque as compared to the outer layers (Geddes, 1977). *Actinomyces* spp are also able to generate high amounts of energy by oxidising lactate when they are in an environment devoid of carbohydrates (van der Hoeven and van der Kieboom, 1990), unlike most streptococcal species. The proportions of Lactobacilli are also higher in carious root dentine than in the overlying plaque (V-702), perhaps due to carious root dentine favouring the propagation of aciduric species such as Lactobacilli in these protected stagnation sites with a reduced access to saliva. Lactobacilli are intrinsically acid tolerant and may only increase in numbers in lesions when there is an extreme selective advantage due to the

predominantly acidic environment in active lesions and other, as yet unknown nutritional factors may also play a role. Such aspects of this type of investigation make the choice of sampling method a crucial factor when comparing studies of Primary Root Caries and these observations together with those relating to the Location and Cavitation of lesions, indicate the need, already referred to, of being able to identify precisely what biopsy has been taken. Investigations on these lines are already planned.

VII CONCLUSIONS

The Hypothesis with which this work has been concerned involves the clinical signs of Primary Root Caries in relation to the Perceived Treatment Needs and the microbiological characteristics. On the basis of clinical signs the presence of a lesion is diagnosed, then clinicians and epidemiologists attempt to classify it as either active or inactive but without having to hand the benefits of a microbiological analysis and yet these must be the foundation on which a reliable diagnosis might be made. From the results of this research the following conclusions are drawn:

- Primary Root Caries lesions with different clinical signs do have different and identifiable microbiological characteristics and, though the Gram-positive pleomorphic rods are the dominant micro-organisms found in all the carious dentine analysed, higher numbers of Mutans streptococci and Lactobacilli have been cultured from carious lesions whose clinical signs were associated with an active caries process than from lesions deemed to be arrested clinically, so these two organisms would seem to be the most reliable microbiological indicators to the activity of a lesion and can be taken as such.

- Significantly higher levels of Mutans streptococci and Lactobacilli are present in Soft caries than Hard caries, in large lesions than small lesions, and in lesions very close to the gingival margin than those remote from it, therefore, the Texture, Size and Location of a lesion are the most reliable clinical criteria of the bacterial activity level of the caries and, of these three, Texture has been shown to

be of greatest significance when the Perceived Treatment Need is being determined.

- The Colour of Primary Root Caries, which has so often been identified as an important indicator of activity, is less reliable than the other three referred to, if only because Black lesions have been found to have significantly higher levels of Mutans streptococci and Lactobacilli than Dark Brown lesions from which they are not always easily distinguished clinically and Black lesions, so commonly equated with arrested caries, are in fact actively carious.

- Since the topical application of the chlorhexidine/thymol tincture described significantly reduces the numbers of Mutans streptococci and Lactobacilli in carious dentine, this indicates a change from an active lesion towards an inactive lesion. Further work needs to be carried out to determine the safest and most effective Chemotherapeutic agent to bring about the virtual sterility of a carious lesion, how it should be employed and how such sterility might be maintained to greatest effect.

- In view of the technical difficulties involved in safely and effectively removing active carious dentine from Primary Root Caries lesions and of restoring such lesions, a most promising line for future research related to this disease would be into its Chemotherapeutic management rather than in its surgical removal, and the restoration of the tooth. Furthermore, equivalent investigations with respect to caries of coronal dentine, notably that related to fissures in the enamel, are indicated. Longitudinal studies of the

Chemotherapeutic management of Primary Root Caries are underway but more will be needed and wider field trials instituted as soon as possible.

- Since the microbiological characteristics of carious dentine on the one hand, and of the overlying plaque on the other, are very different, it is essential in every investigation to ensure that any biopsy taken is precisely identified and does not include a mixture of samples from the dentine itself, the overlying plaque and the plaque on non-carious enamel or dentine surfaces. Furthermore, the precise Location from which a biopsy is taken must be able to be identified with some precision with reference to the lesion itself, its depth and the gingivae. This is now feasible by means of the analysis of replicas using a co-ordinate measuring machine and is currently being investigated.

- In view of the severe limitations imposed upon epidemiologists in obtaining data relating to Primary Root Caries, any such data obtained to date needs to be viewed with caution in view of this research, not least the unreliability of Colour as a criterion and the importance of Texture, which epidemiological examinations would find difficult to determine. New clinical criteria will, therefore, need to be universally agreed which include Location, Size and Cavitation which have been shown to be significantly correlated with Texture and much less emphasis should be laid on Colour than hitherto.

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